

Mimics of Complex Carbohydrates Recognized by Receptors

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Carbohydrates are found ubiquitously in nature, as part of natural products, as sources of metabolic energy, and as key components for various intercellular recognition processes, including infection, inflammation, metastasis, differentiation, development, and regulation of signaling. Although carbohydrates play key roles in these vital biological recognition processes and in development of diseases, the detailed mechanisms of these events are poorly understood. As such, carbohydrates remain the least exploited among the three major classes of biomolecules. Chemists consider complex carbohydrates difficult to synthesize and manipulate, and their biological functions are often not well defined. Biologists do not have useful tools to study carbohydrates. There is no amplification method available for carbohydrates to facilitate structure analysis. There is no machine available for the synthesis of oligosaccharides to exploit their functions. In addition, there is no general method available for the preparation of glycoproteins with well-defined carbohydrates to study the roles of carbohydrates in glycoprotein structure and function. Medicinal chemists consider complex carbohydrates as an uninteresting class of molecules for drug development, since carbohydrates are usually too complex for process development and too hydrophilic to have good bioavailability, and they are generally orally inactive and unstable. Perhaps the most fundamental problem is that carbohydrates bind their receptors with weak affinity, usually with dissociation constants in the millimolar range, presumably due to the lack of hydrophobic and charged groups on carbohydrates.^{1,2} Although nature uses multivalency to improve the affinity and specificity in carbohydrate–receptor interaction on cell surfaces, the multivalent approach to drug development still represents a significant challenge.^{3–9}

Chi-Huey Wong was born in Taiwan in 1948. He received his B.S. and M.S. degrees from the National Taiwan University and his Ph.D. in chemistry (with George M. Whitesides) from the Massachusetts Institute of Technology in 1982. After a year of postdoctoral work at Harvard University (with Whitesides), he taught at Texas A&M University for six years and then moved to The Scripps Research Institute as Professor and Ernest W. Hahn Chair in Chemistry in 1989. His research interests include carbohydrate recognition, enzymes in organic synthesis, enzyme inhibition and reaction mechanisms.

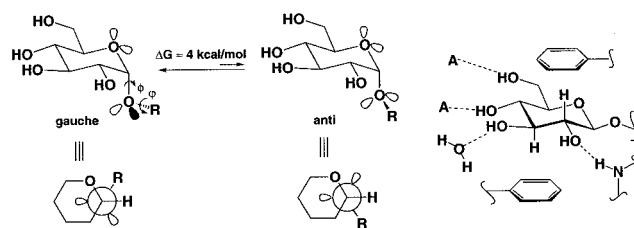


FIGURE 1. (Left) The conformation of oligosaccharides in solution is mainly governed by the *exo*-anomeric and steric effects. The *exo*-anomeric effect is the antiperiplanar alignment of a lone pair orbital of the glycosidic oxygen and the bond between the ring oxygen and anomeric carbon. As a consequence, the aglycon group (R) tends to be away from the sugar ring substituents as indicated in the gauche form to reduce the steric hindrance, and the two glycosidic torsion angles φ and ϕ are thus defined. (Right) The interaction of carbohydrates with proteins involves multiple H-bonding interactions with water and protein side chains and aromatic sugar-ring C–H interactions. In many cases, the bound conformation is not very different from the solution conformation.

It is, however, important to study carbohydrate-mediated biological processes and to investigate their mechanisms of action, as understanding the mechanism of carbohydrate function may lead to the development of carbohydrate-based therapeutics. Recent advances in glycobiology and glycochemistry have helped solve some of the problems associated with carbohydrate research, and methods for the synthesis of some complex carbohydrates on a large scale for drug development are now available.⁴ Current efforts are, however, directed toward the development of small molecules to mimic the structure and function of complex carbohydrates, with the hope that more active, more stable, and perhaps orally active small molecules that are easily synthesized can be developed. This Account focuses on some of the most recent efforts in this regard and concentrates on the development of small molecules that mimic the sialyl Lewis x tetrasaccharide (sLe^x) recognized by selectins, the transition states in glycosidase- and glycosyltransferase-catalyzed reactions and the aminoglycoside antibiotics binding to RNA.

Mimics of Sialyl Lewis x Recognized by Selectins

Carbohydrate–protein interactions contain complex H-bonding networks involving water molecules and the functional groups of sugars, and hydrophobic interactions between the C–H groups of sugars and protein side chains such as aromatic groups (Figure 1).² The conformation of an oligosaccharide bound to a receptor is, in many cases, not very different from that in solution, which is mainly governed by the *exo*-anomeric effects (Figure 1).¹ A case study presented in this Account is the interaction of sLe^x tetrasaccharide ligand with a class of calcium-dependent lectins, including E-, P-, and L-selectins (Figure 2).⁴ The selectin–carbohydrate interaction is involved in

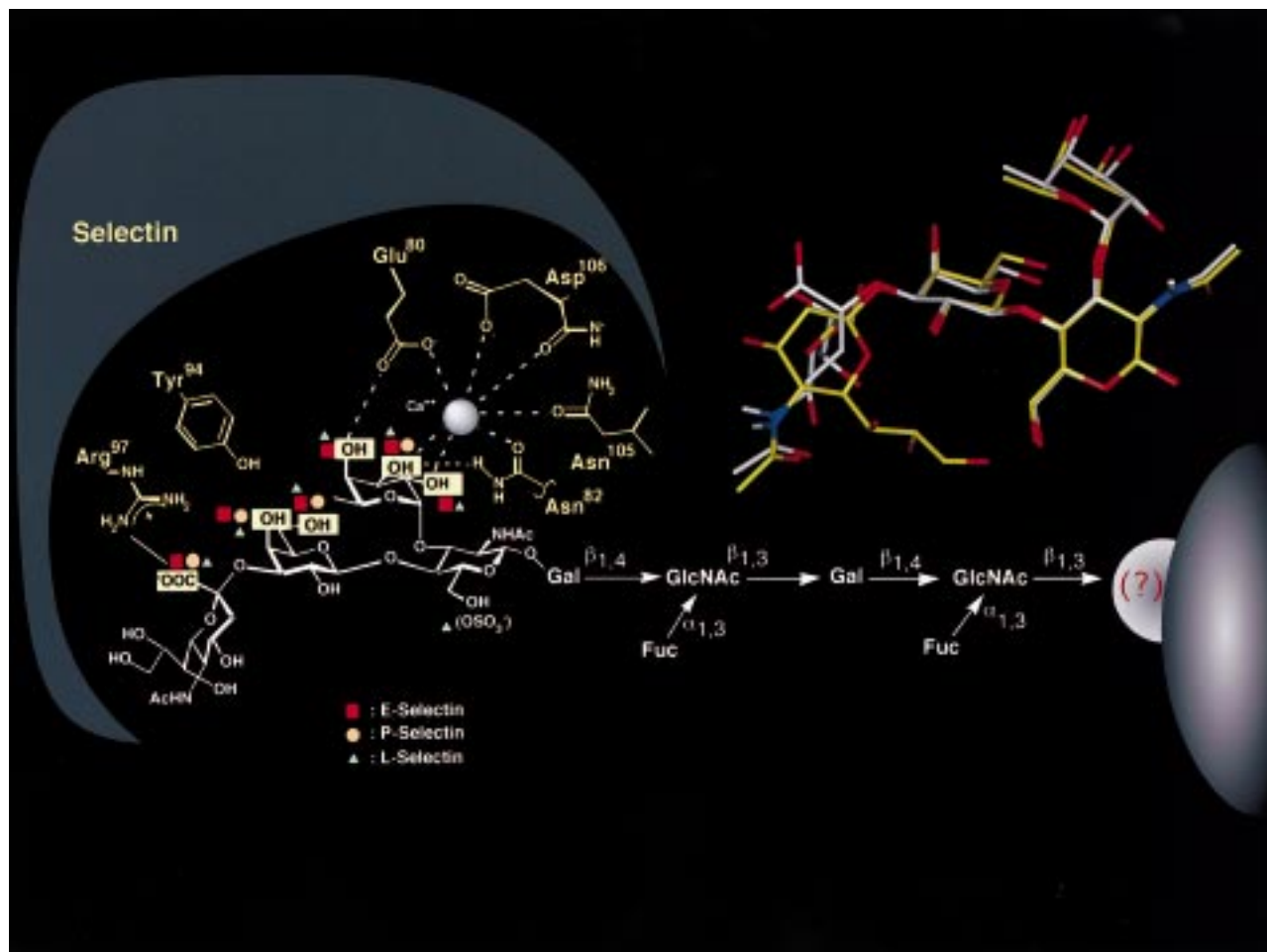


FIGURE 2. Essential functional groups of sialyl Lewis x interacting with selectin side chains and conformations of sLe^x bound to E-, P-, and L-selectins are indicated. Those shown as red squares are recognized by E-selectin, those shown as orange circles are recognized by P-selectin, and those shown as green triangles are recognized by L-selectin. Regarding the bound conformation, the one in yellow is recognized by E- and P-selectins and that in white is close to the solution conformation and recognized by L-selectin. The precise carbohydrate sequence between sLe^x and protein is not known. The main difference between the bound conformations is the orientation of the carboxylate group.

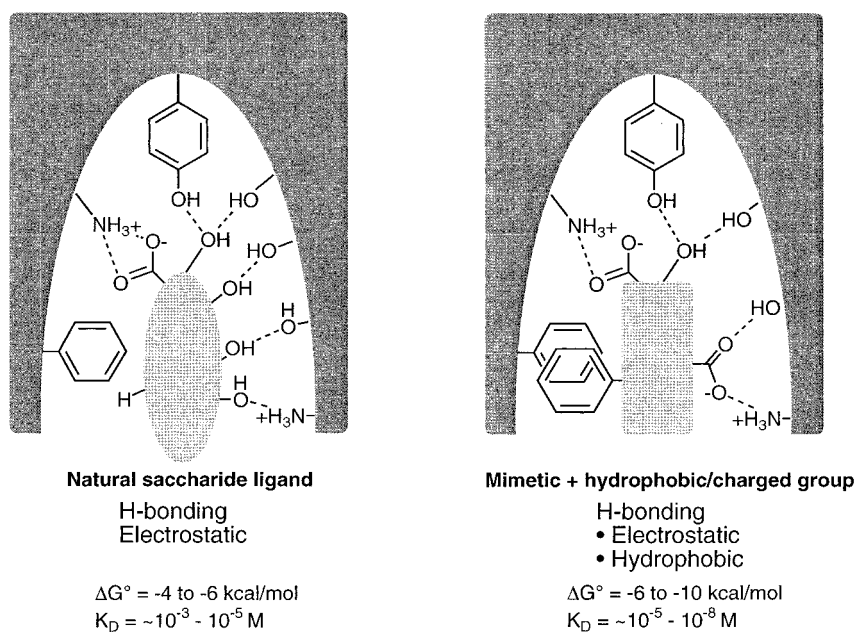


FIGURE 3. Strategy for the development of complex carbohydrate mimetics. An additional lipophilic or charged group is incorporated in the scaffold containing the essential groups for binding to further increase the affinity.

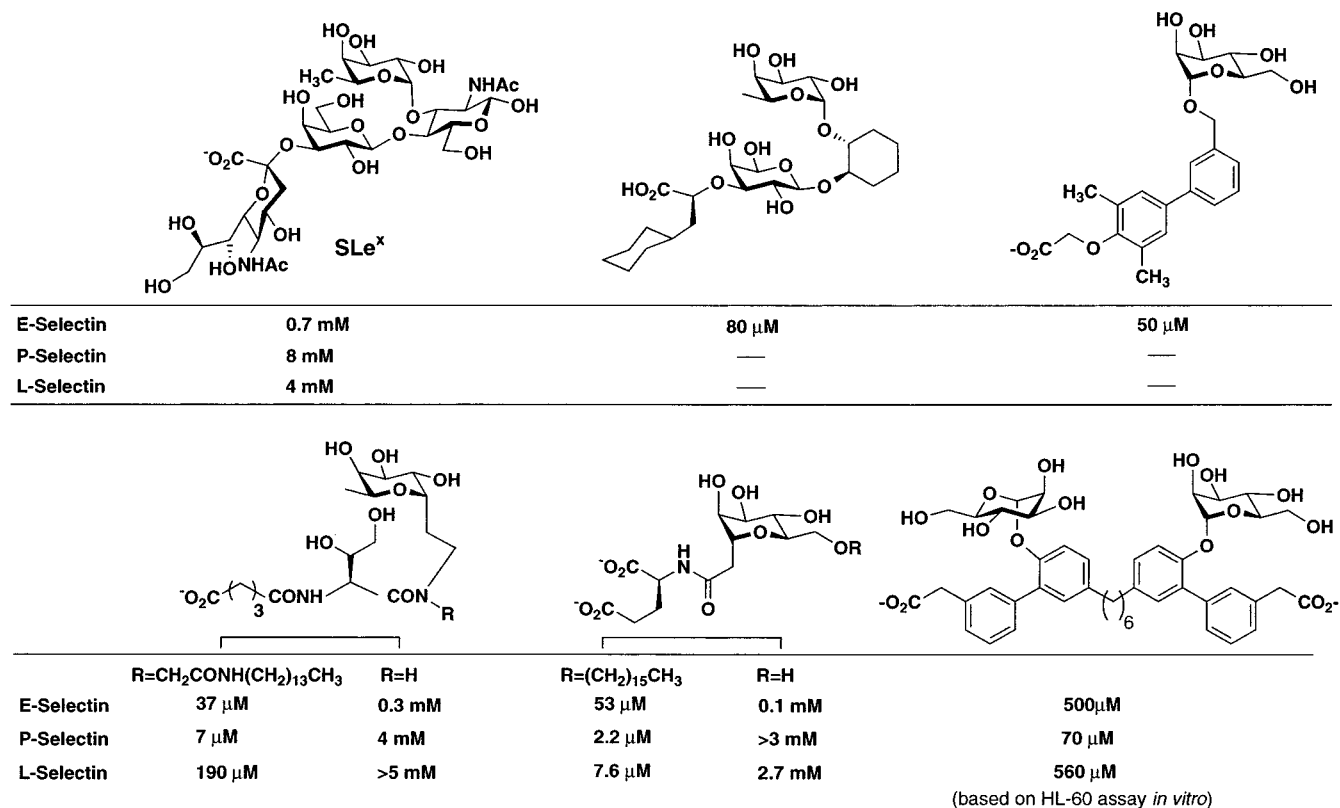


FIGURE 4. Representative mimetics of sialyl Lewis x. For the top middle one, see: Kolb, H.-C.; Ernst, B. *Chem. Eur. J.* **1997**, *3*, 1571. For the bottom right one, see: Kogan, T. P.; et al. *J. Med. Chem.* **1998**, *41*, 1099. For the others see ref 4.

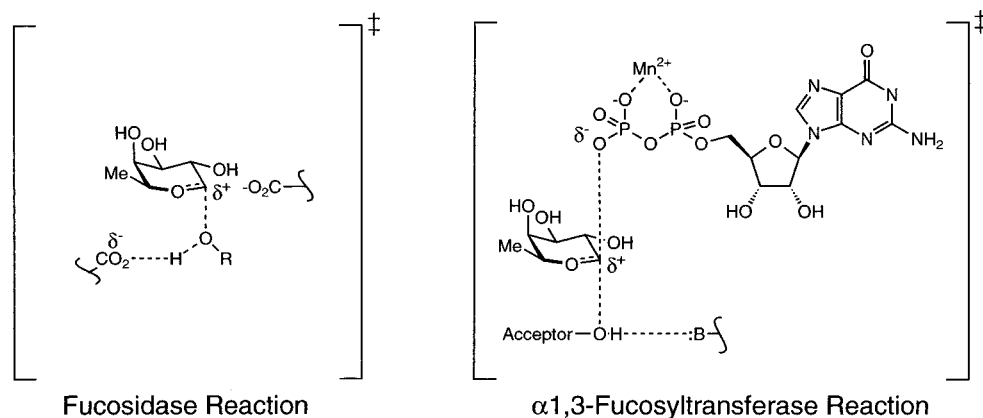


FIGURE 5. Postulated mechanisms for glycosidase and glycosyltransferase reactions as illustrated by a fucosidase and a fucosyltransferase reaction.

inflammatory reactions and, in certain cases, metastasis.⁴ A common process leading to inflammation is when a tissue is injured or stimulated by inflammatory factors such as bacterial lipopolysaccharides, leukotriene B₄, and other toxins, cytokines such as tumor necrosis factor (TNF) and interleukin-1 (IL-1) along with nitric oxide are released to signal the display of P-selectin (occurring in 20–30 min) and E-selectin (in ~60 min) on the surface of endothelial cells. The rolling leukocytes then adhere to the surface of endothelial cells through the multivalent interaction between the selectin and the tetrasaccharide sLe^x, which is the nonreducing terminal moiety of a glycoprotein

expressed on the surface of leukocytes. This carbohydrate–protein interaction is followed by the tight protein–protein interaction between immunoglobulin superfamily molecules, such as intercellular adhesion molecule-1 (ICAM-1) or vascular adhesion molecule-1 (VCAM-1), on the endothelium with integrins on the leukocyte cell surface, which leads to the extravasation of leukocytes to the site of injury. Blocking this protein–carbohydrate interaction has been considered to be a new strategy for the treatment of acute and chronic inflammatory diseases and postsurgical reperfusion injury. Indeed, it has been shown that a synthetic sLe^x derivative is effective for the

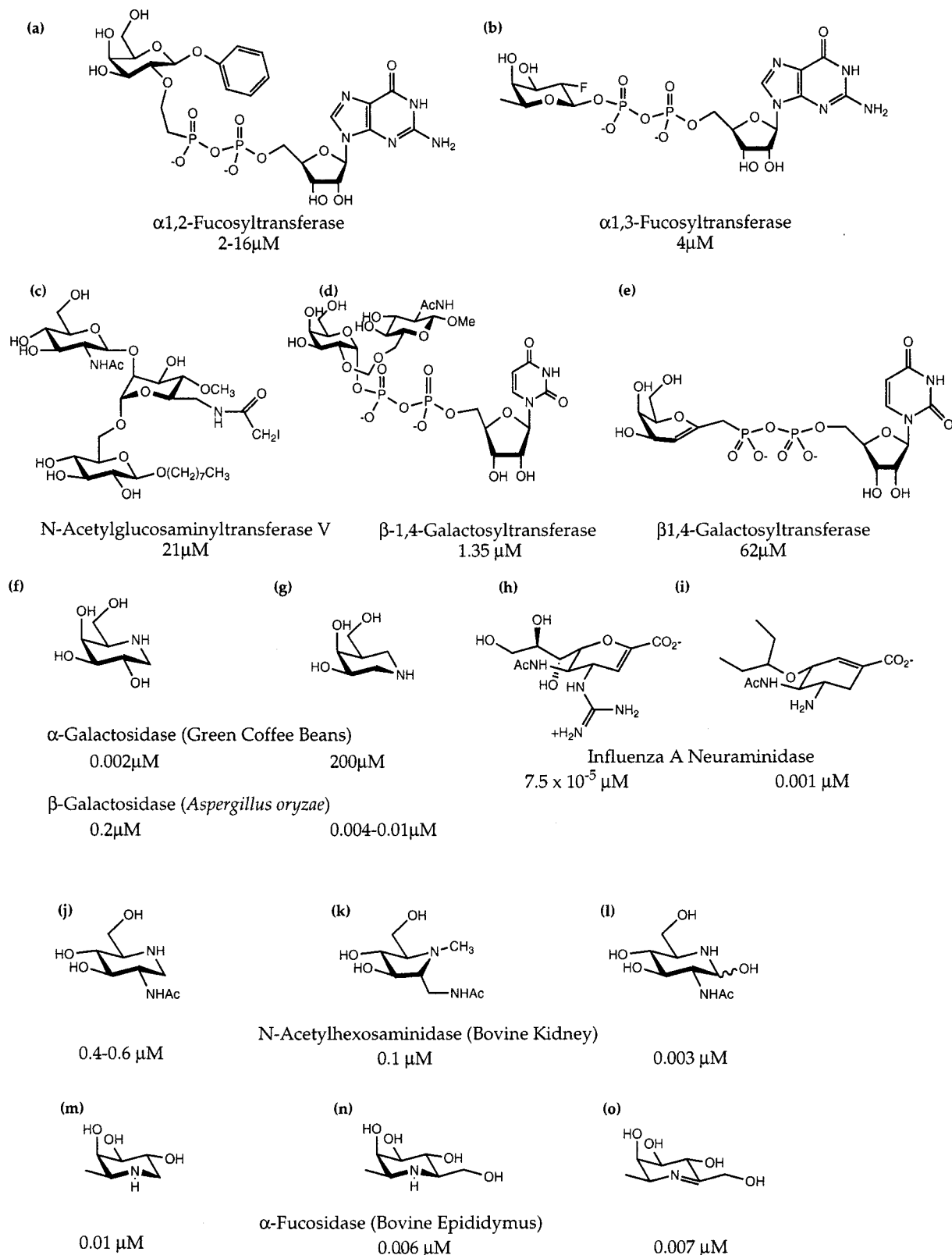


FIGURE 6. Representative inhibitors of glycosidases and glycosyltransferases (K_i in μ M). (a) See: Palcic, M. M.; Heerze, L. D.; Srivastava, O. P.; Hindsgaul, O. *J. Biol. Chem.* **1989**, *264*, 17174. (b) See ref 20. (c) See: Lu, P.-P.; Hindsgaul, O.; Campston, C. A.; Palcic, M. M. *Bioorg. Med. Chem.* **1996**, *4*, 2011. (d) See: Hashimoto, H.; Endo, T.; Kajihara, Y. *J. Org. Chem.* **1997**, *62*, 1914. (e) See: Schmidt, R. R.; Frische, K. *Bioorg. Med. Chem. Lett.* **1993**, *3*, 1747. (f) See ref 17. (g) See refs 18 and 19. (h) See ref 24. (i) See ref 25. (j) See: Kappes, E.; Legler, G. *J. Carbohydr. Chem.* **1989**, *8*, 371. Kajimoto, T.; Liu, K.; Pederson, R. L.; Zhong, Z.; Ichikawa, Y.; Porco, J. A.; Wong, C.-H. *J. Am. Chem. Soc.* **1991**, *113*, 6187. (k) See: Takabiashi, M.; Kanie, O.; Wong, C.-H. Unpublished results. (l) See reference given for (j). (m, n) See: Fleet, G. W. J.; Namguong, S. K.; Barker, C.; Baines, S.; Jacob, G. S.; Winchester, B. *Tetrahedron Lett.* **1989**, *30*, 4439. Fleet, G. W. J.; Shaw, A. N.; Evans, S. V.; Fellows, L. E. *Chem. Commun.* **1985**, 841. (o) See ref 23.

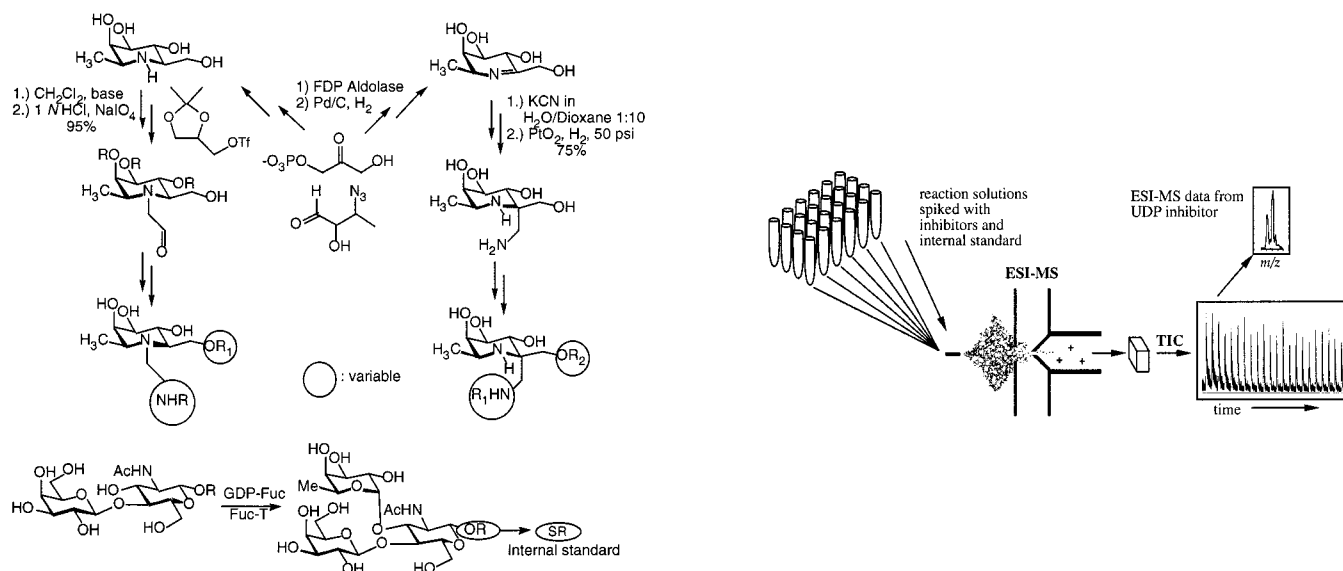


FIGURE 7. (Left) A representative library approach to the discovery of selective inhibitors of fucosidases and fucosyltransferases. A common core mimicking the transition state of the glycosyltransfer moiety is linked to other group(s) to mimic the leaving group (in glycosidase reactions) or the acceptor and the nucleoside leaving groups (in glycosyltransferase reactions). (Right) Use of electrospray mass spectrometry in rapid screening of inhibitors. The thioglycoside is used as an internal standard. Fuc = fucose and FucT = fucosyltransferase.

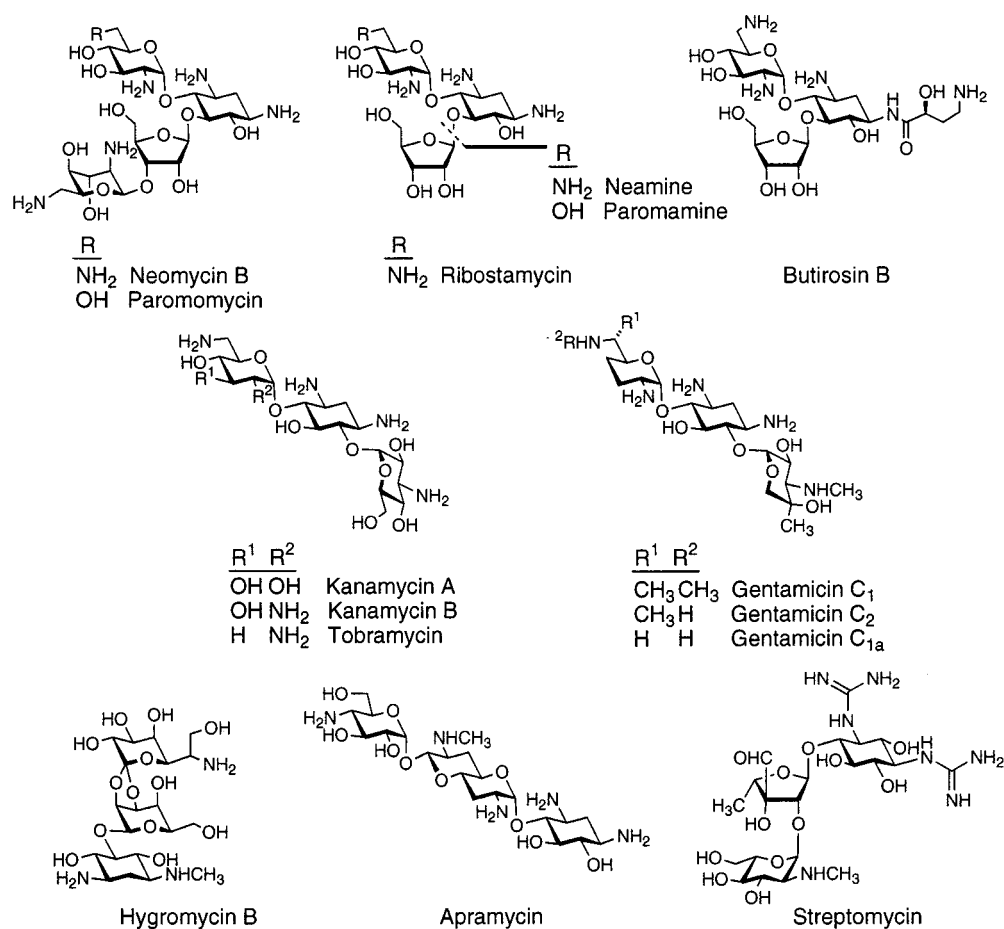


FIGURE 8. Representative structures of aminoglycoside antibiotics.

treatment of reperfusion injury during heart surgery.¹⁰ It, however, requires the use of a high dose of sLe^x to achieve the desirable inhibition activity as sLe^x binds the selectins with dissociation constants in the millimolar range.

Through structure–function relationship studies, the essential groups of sLe^x recognized by selectins have been identified.¹¹ The conformations of sLe^x bound to the selectins have also been determined with the use of the

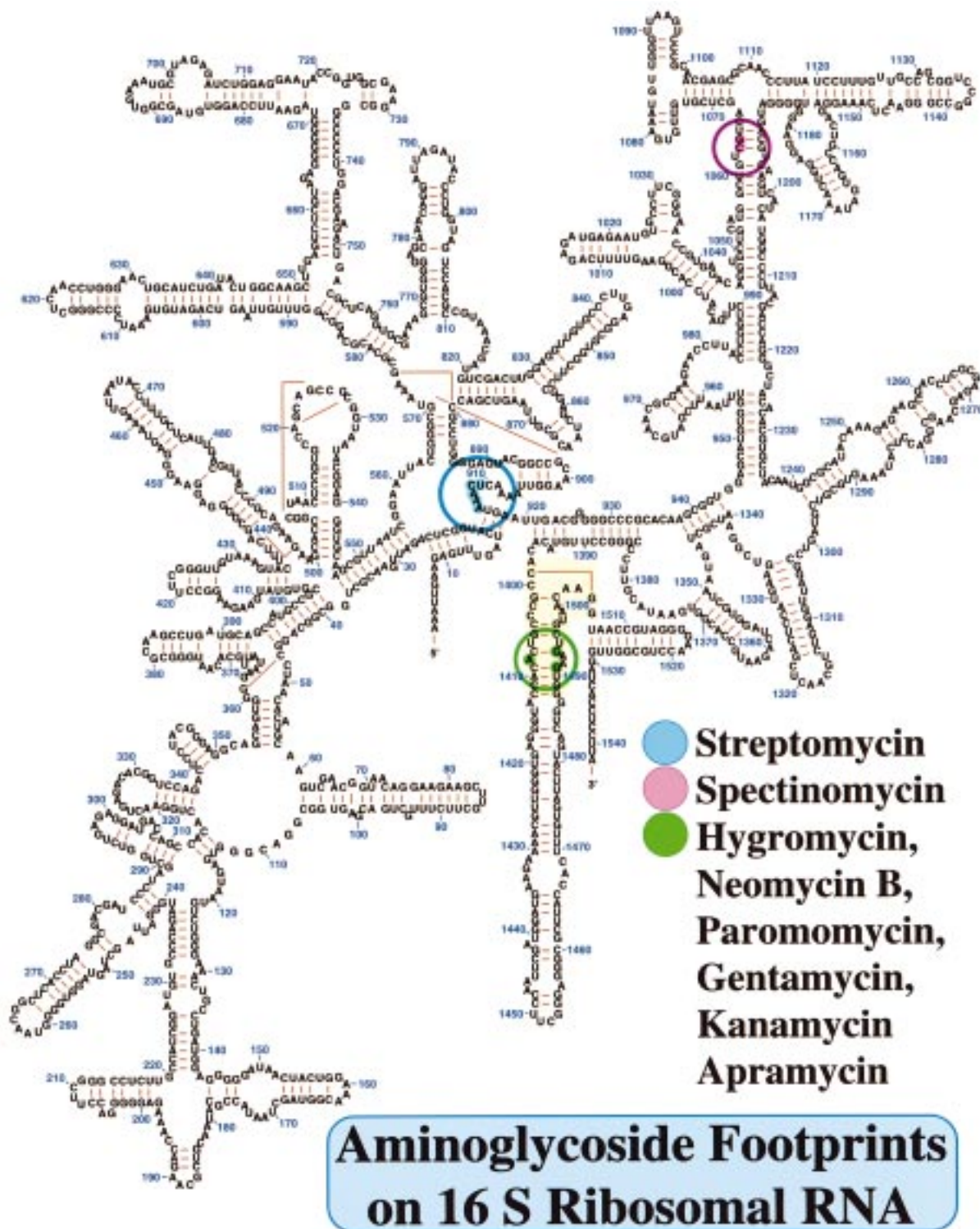


FIGURE 9. Structure of the bacterial 16S RNA A site and the site for aminoglycoside antibiotic binding.

transfer NOE technique^{12,13} (Figure 2). These two pieces of information have provided a basis for the design of sLe^x mimetics as inhibitors of selectins, and to date many small molecules with affinity equal to or greater than that of sLe^x have been prepared.⁴ One of the most effective strategies for the development of sLe^x mimetics is to incorporate an additional lipophilic or charged group in

a scaffold that contains the essential functional groups for recognition as illustrated in Figure 3, if the new lipophilic (or charged) group interacts favorably with a lipophilic (or charged) residue in the receptor. This change will result in a substantial increase in affinity due to the entropic consequences of the hydrophobic effect (or the electrostatic interaction). Identification of such an additional

group, however, requires structural information on the selectin–carbohydrate complex, which is still not available at this time, though the X-ray structure of E-selectin has been determined.¹⁴ A combinatorial or library approach to the identification of such groups becomes an alternative option. In any event, small molecules with inhibitory potency in the low micromolar range have been discovered (see Figure 4 for representative examples), and constrained cyclic structures based on the newly discovered sLe^x mimetics with even better biological activity may be developed eventually.

Mimics of Transition States in Glycosidase- and Glycosyltransferase-Catalyzed Reactions

Glycosidases and glycosyltransferases are important catalysts for the processing and synthesis of carbohydrates, including polysaccharides, glycolipids, and glycoproteins. Inhibition of these enzymes associated with disease states or metabolic disorders is another strategy for carbohydrate-based therapy.¹⁵ The mechanisms of glycosidases, including inverting and retaining enzymes, have been well studied,^{16–17} and means for inhibition of these enzymes have been developed, including the use of natural products and synthetic inhibitors.^{18–20} Glycosyltransferase-catalyzed reactions are thought to proceed through a transition state similar to that of the glycosidase reactions, with a partially positively charged half-chair structure and sp² character developed at the anomeric center²¹ (Figure 5). The glycosidic bond of the sugar nucleotide in the transferase reaction was shown to be broken prior to the nucleophilic attack by the incoming acceptor, as illustrated in the human α -1,3-fucosyltransferase V and β -1,4-galactosyltransferase reactions.^{21–22} Whether this S_N1-like mechanism is general in glycosyltransferase reactions, however, remains to be established. On the basis of these mechanistic rationales, many potent inhibitors of glycosidases and glycosyltransferases have been developed. Of particular interest are inhibitors that contain a positively charged endocyclic or exocyclic nitrogen to mimic the transition-state structure of the enzymatic reactions (Figure 6). This new electrostatic interaction developed in the glycosyl transfer reaction contributes significantly to transition-state binding. These inhibitors, however, often exhibit little selectivity, with some exceptions, as they usually do not interact with the leaving group binding site and further modification is necessary to improve selectivity. A useful approach is to incorporate additional groups to the heterocycles or aminocarbocycles to occupy the binding site for the leaving group (in the case of glycosidases) or the binding site for the leaving group and the acceptor group (in the case of glycosyltransferases) (Figure 7). This additional group can be found by rational design or by a library approach combined with a fast inhibition analysis using techniques such as electrospray mass spectrometry or other high throughput analysis.^{23,24}

It is noted that mimicking the transition state of an enzymatic reaction has been an effective strategy for the

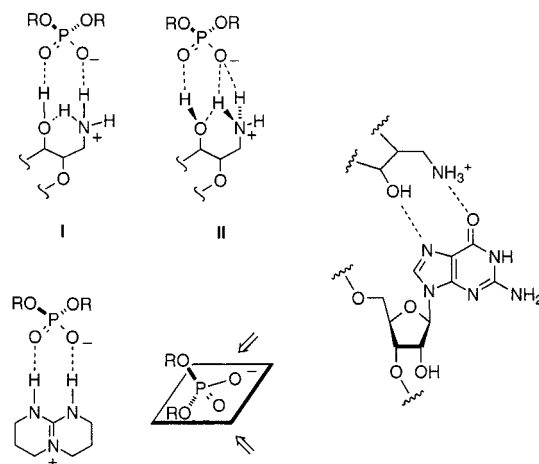


FIGURE 10. The β -1,3-hydroxyamine motif found in aminoglycoside antibiotics interacts more strongly than does the standard cyclic guanidine group with phosphodiester and the Hougsteen face of guanine. The stronger interaction is perhaps due to the presence of localized charge and multiple H-bonds formed above and below the OPO plane as shown in II.

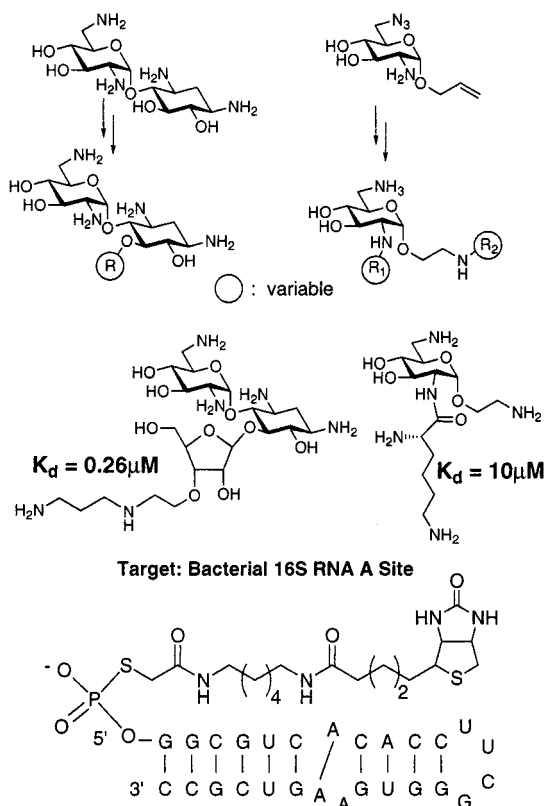


FIGURE 11. A library approach to the discovery of aminoglycoside mimetics as antibiotics using the β -1,3-hydroxyamine motif or the neamine moiety as the core. Both Petri dish assay by the Kirby–Dauer disk method and surface plasmon resonance assay using a biotinylated RNA sequence of the bacterial 16S A site are used to select new antibiotics. Two representative aminoglycoside mimetics with their K_d values are illustrated.

development of potent enzyme inhibitors, including inhibitors of glycosidases such as influenza neuraminidase.^{25–26} Development of potent and specific inhibitors of glycosyltransferases, however, still represents a significant challenge.

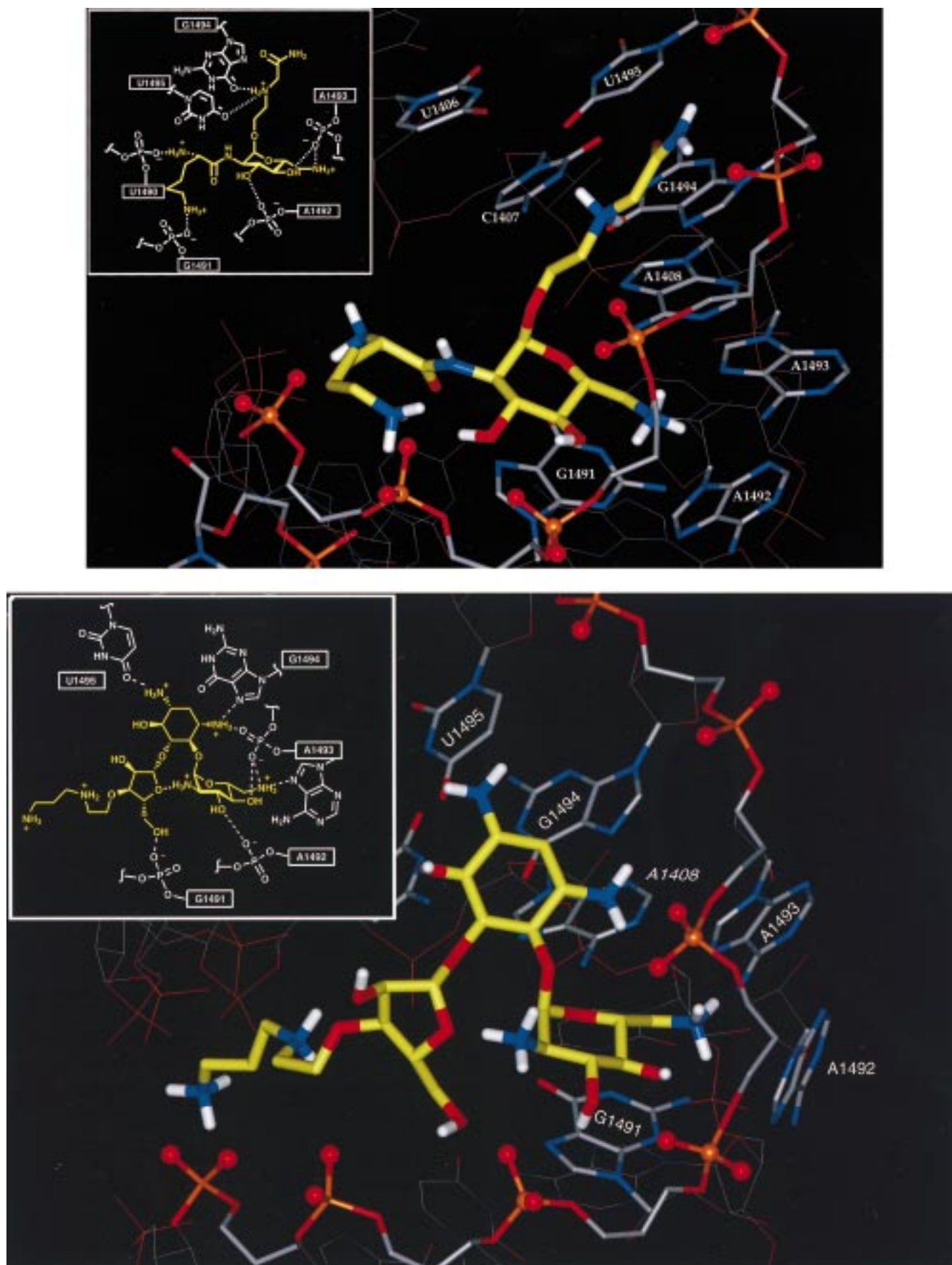


FIGURE 12. Computer modeling of two representative newly discovered antibiotics interacting with a 16S RNA A site model. The modeling is based on the structure of paromomycin complexed with the 16S RNA A site model.³¹

Mimics of Aminoglycoside Antibiotics Binding to RNA

The antibiotic activities of many naturally occurring aminoglycosides (Figure 8) arise from their interaction with bacterial RNA, especially the 16S A site (Figure 9), resulting in truncation and miscoding in protein biosynthesis.²⁷ Aminoglycoside antibiotics are orally inactive and often administered by injection or topical application. In addition, most of the antibiotics have experienced drug resistance due to modification of the antibiotics (via acetylation and phosphorylation) or the RNA by the enzymes of targeted microorganisms. An interesting feature of aminoglycoside antibiotics is that they often contain a *trans*-1,3-hydroxyamine or *cis*-1,3-diamine motif. A recent model study by NMR²⁸ indicates that the glyco-type 1,3-hydroxyamine interacts, through multiple H-bonds, with phosphodiester and the Hougsteen face of guanine more strongly than does the standard cyclic guanidine group (Figure 10). This finding has led to the discovery of new aminoglycoside mimetics containing either one or both of these recognition motifs that recognize RNA (Figure 11), as illustrated in the binding analysis of synthetic libraries using surface plasmon resonance.^{29,30} Some of the mimetics exhibit strong antibiotic activities. Computer modeling also suggests that the mimetics bind to RNA in a manner similar to that of the natural products,³¹ with the hydroxyamine motif interacting with the phosphodiester group and the Hougsteen face of guanine (Figure 12). This approach points to a new direction for the development of potent sequence-specific RNA binders, a strategy that might be extended to study the problem of DNA recognition. Incorporation of a reactive group in the mimetics may generate a sequence-specific irreversible inhibitor. Furthermore, understanding the binding stoichiometry based on the surface plasmon resonance analysis will lead to the development of multivalent aminoglycoside mimetics to further improve affinity and specificity.

Conclusions and Future Prospects

The three case studies cited here illustrate how the complexity and weak affinity in carbohydrate-receptor interactions can be addressed with the use of structurally simplified carbohydrate mimetics that contain the essential functional groups of the parent ligand. Incorporation of additional hydrophobic and/or charged groups in the mimetics, however, represents a key feature in the development of mimetics with high affinity and specificity. This approach is consistent with findings from structural studies of a limited number of carbohydrate-receptor complexes, where, in addition to the complex H-bonding network, the sugar face C-H group stacking with aromatic residues contributes significantly to the overall binding affinity and stereospecificity. Introducing a new hydrophobic group on the sugar ligand or its mimetic may enhance hydrophobic interactions, thus improving affinity and specificity. It is hoped that the strategies illustrated here are generally applicable to other carbohydrate-

mediated biological recognition and are useful for the development of carbohydrate-based therapeutics.

References

- (1) Lemieux, R. U. How Water Provides the Impetus for Molecular Recognition in Aqueous Solution. *Acc. Chem. Res.* **1996**, *29*, 373.
- (2) Quijcho, F.-A. Protein-carbohydrate interactions: basic molecular features. *Pure Appl. Chem.* **1989**, *61*, 1293.
- (3) Lee, Y. C.; Lee, R. T. Carbohydrate-Protein Interactions: Basis of Glycobiology. *Acc. Chem. Res.* **1995**, *28*, 321.
- (4) Simanek, E. E.; McGarvey, G. J.; Jablonowski, J. A.; Wong, C.-H. Selectin-Carbohydrate Interactions: From Natural Ligands to Designed Mimics. *Chem. Rev.* **1998**, *98*, 833.
- (5) Kiessling, L. L.; Pohl, N. Strength in numbers: non-natural polyvalent carbohydrate derivatives. *Chem. Biol.* **1996**, *3*, 71.
- (6) DeFrees, S. A.; Philips, L.; Guo, L.; Zalipsky, S. Sialyl Lewis x Liposomes as a Multivalent Ligand and Inhibitor of E-Selectin-Mediated Cellular Adhesion. *J. Am. Chem. Soc.* **1996**, *118*, 1601.
- (7) Spevak, W.; Foxall, C.; Charych, D. H.; Dasgupta, F.; Nagy, J. O. Carbohydrates in an Acidic Multivalent Assembly: Nanomolar P-Selectin Inhibitors. *J. Med. Chem.* **1996**, *39*, 1018.
- (8) Sigal, G. B.; Mammen, M.; Dahmann, G.; Whitesides, G. M. Polyacrylamides Bearing Pendant α -Sialoside Groups Strongly Inhibit Agglutination of Erythrocytes by Influenza Virus: The Strong Inhibition Reflects Enhanced Binding through Cooperative Polyvalent Interactions. *J. Am. Chem. Soc.* **1996**, *118*, 3789.
- (9) Kamitakahara, H.; Suzuki, T.; Nishigori, N.; Suzuki, Y.; Kanie, O.; Wong, C.-H. A Lysoganglioside/Poly-L-glutamic Acid Conjugate as a Picomolar Inhibitor of Influenza Hemagglutinin. *Angew. Chem., Int. Ed. Engl.* **1998**, *37*, 1524.
- (10) Murohara, T.; Marigiotta, J.; Phillips, L. M.; Paulson, J. C.; DeFrees, S.; Zalipsky, S.; Guo, L. S.; Lefer, A. M. Cardioprotection by liposome-conjugated sialyl Lewis x-oligosaccharide in myocardial ischemia and reperfusion injury. *Cardiovas. Res.* **1995**, *30*, 965.
- (11) Bradley, N. K.; Kiso, M.; Abbas, S.; Nikrad, P.; Srivastava, O.; Foxall, D.; Oda, Y.; Hasegawa, A. Structure-function studies on selectin carbohydrate ligands. Modifications to fucose, sialic acid and sulfate as a sialic acid replacement. *Glycobiology* **1993**, *3*, 663 and citations in ref 4.
- (12) Scheffler, K.; Ernst, B.; Katopodis, A.; Magnani, J. L.; Wang, W. T.; Weisemann, R.; Petrs, T. Determination of the bioactive conformation of the carbohydrate ligand in the E-selectin/sialyl Lewis complex. *Angew. Chem., Int. Ed. Engl.* **1995**, *34*, 1841.
- (13) Poppe, L.; Brown, G. S.; Philo, J. S.; Nikrad, P. V.; Shah, B. H. Conformation of sLe^x Tetrasaccharide, Free in Solution and Bound to E-, P-, and L-Selectin. *J. Am. Chem. Soc.* **1997**, *119*, 1727.
- (14) Graves, B. J.; Crowther, R. L.; Chandran, C.; Rumberger, J. M.; Li, S.; Huang, K.-S.; Presky, D. H.; Famittetti, P. C.; Wolitzky, B. A.; Burns, D. K. *Nature* **1994**, *367*, 532.
- (15) Sears, P.; Wong, C.-H. Mechanism-based inhibition of carbohydrate-mediated biological recognitions. *Chem. Commun.* **1998**, 1161.
- (16) Sinnott, M. L. Catalytic mechanism of enzymic glycosyl transfer. *Chem. Rev.* **1990**, *90*, 1171.

- (17) Withers, S. G.; Street, I. P. Identification of a covalent α -D-glucopyranosyl enzyme intermediate formed on a β -glucosidase. *J. Am. Chem. Soc.* **1988**, *110*, 8551.
- (18) Ganem, B. Inhibitors of Carbohydrate-Processing Enzymes: Design and Synthesis of Sugar-Shaped Heterocycles. *Acc. Chem. Res.* **1996**, *29*, 340.
- (19) Bols, M. 1-aza sugars, apparent transition state analogs of equatorial glycoside formation/cleavage. *Acc. Chem. Res.* **1998**, *31*, 1.
- (20) Ichikawa, Y.; Igarashi, Y.; Ichikawa, M.; Suhara, Y. 1-N-Imino Sugars: Potent and Selective Inhibitors of β -Glycosidases. *J. Am. Chem. Soc.* **1998**, *120*, 3007.
- (21) Murray, B. W.; Wittmann, V.; Burkart, M.; Hung, S.-C.; Wong, C.-H. Mechanism of Human α -1,3-Fucosyltransferase V: Glycosidic Cleavage Occurs Prior to Nucleophilic Attack. *Biochemistry* **1997**, *36*, 823.
- (22) Hayashi, T.; Murray, B. W.; Wang, R.; Wong, C.-H. A Chemoenzymatic Synthesis of UDP-(2-deoxy-2-fluoro)-galactose and Evaluation of its Interaction with Galactosyltransferase. *Bioorg. Med. Chem.* **1997**, *4*, 497.
- (23) Wu, J.; Takayama, S.; Wong, C.-H.; Siuzdak, G. Quantitative electrospray mass spectrometry for the rapid assay of enzyme inhibitors. *Chem. Biol.* **1997**, *4*, 653.
- (24) Takayama, S.; Martin, R.; Wu, J.; Laslo, K.; Siuzdak, G.; Wong, C.-H. Chemoenzymatic Preparation of Novel Cyclic Imine Sugars and Rapid Biological Activity Evaluation Using Electrospray Mass Spectrometry and Kinetic Analysis. *J. Am. Chem. Soc.* **1997**, *119*, 8146.
- (25) Von Itzstein, M.; Wu, W.-Y.; Kok, G. B.; Pegg, M. S.; Dyason, J. C.; Jin, D.; Van Phan, T.; Smythe, M. L.; White, H. F.; Oliver, S. W.; Colman, P. M.; Varghese, J. N.; Ryan, D. M.; Woods, J. M.; Bethell, R. C.; Hotham, V. J.; Cameron, J. M.; Penn, C. R. Rational design of potent sialidase-based inhibitors of influenza virus replication. *Nature* **1993**, *363*, 418.
- (26) Kim, C. U.; Lew, W.; Williams, M. A.; Liu, H.; Zhang, L.; Swaminathan, S.; Bischofberger, N.; Chen, M. S.; Mendel, D. B.; Tai, C. Y.; Laver, W. G.; Stevens, R. C. Influenza Neuraminidase Inhibitors Possessing a Novel Hydrophobic Interaction in the Enzyme Active Site: Design, Synthesis, and Structural Analysis of Carbocyclic Sialic Acid Analogs with Potent Anti-Influenza Activity. *J. Am. Chem. Soc.* **1997**, *119*, 681.
- (27) von Ahsen, U.; Noller, H. F. Footprinting the sites of interaction of antibiotics with catalytic group I intron RNA. *Science* **1993**, *260*, 1506.
- (28) Hendrix, M.; Alper, P. B.; Priestley, E. S.; Wong, C.-H. Hydroxyamines as a New Motif for the Molecular Recognition of Phosphodiester: Implications for Aminoglycoside-RNA Interactions. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 95.
- (29) Alper, P. B.; Hendrix, M.; Sears, P.; Wong, C.-H. Probing the Specificity of Aminoglycoside-Ribosomal RNA Interactions with Designed Synthetic Analogs. *J. Am. Chem. Soc.* **1998**, *120*, 1965.
- (30) Wong, C.-H.; Hendrix, M.; Manning, D. D.; Rosenbohm, C.; Greenberg, W. A. A library approach to the discovery of small molecules that recognize RNA: Use of a 1,3-hydroxyamine motif as core. *J. Am. Chem. Soc.* **1998**, *120*, 8319.
- (31) Fourmy, D.; Recht, M. I.; Blanchard, S. C.; Puglisi, J. D. Structure of the A site of Escherichia coli 16S ribosomal RNA complexed with an aminoglycoside antibiotic. *Science* **1996**, *274*, 1367.

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