Supporting Information

Shah *et al.* 10.1073/pnas.0802206105

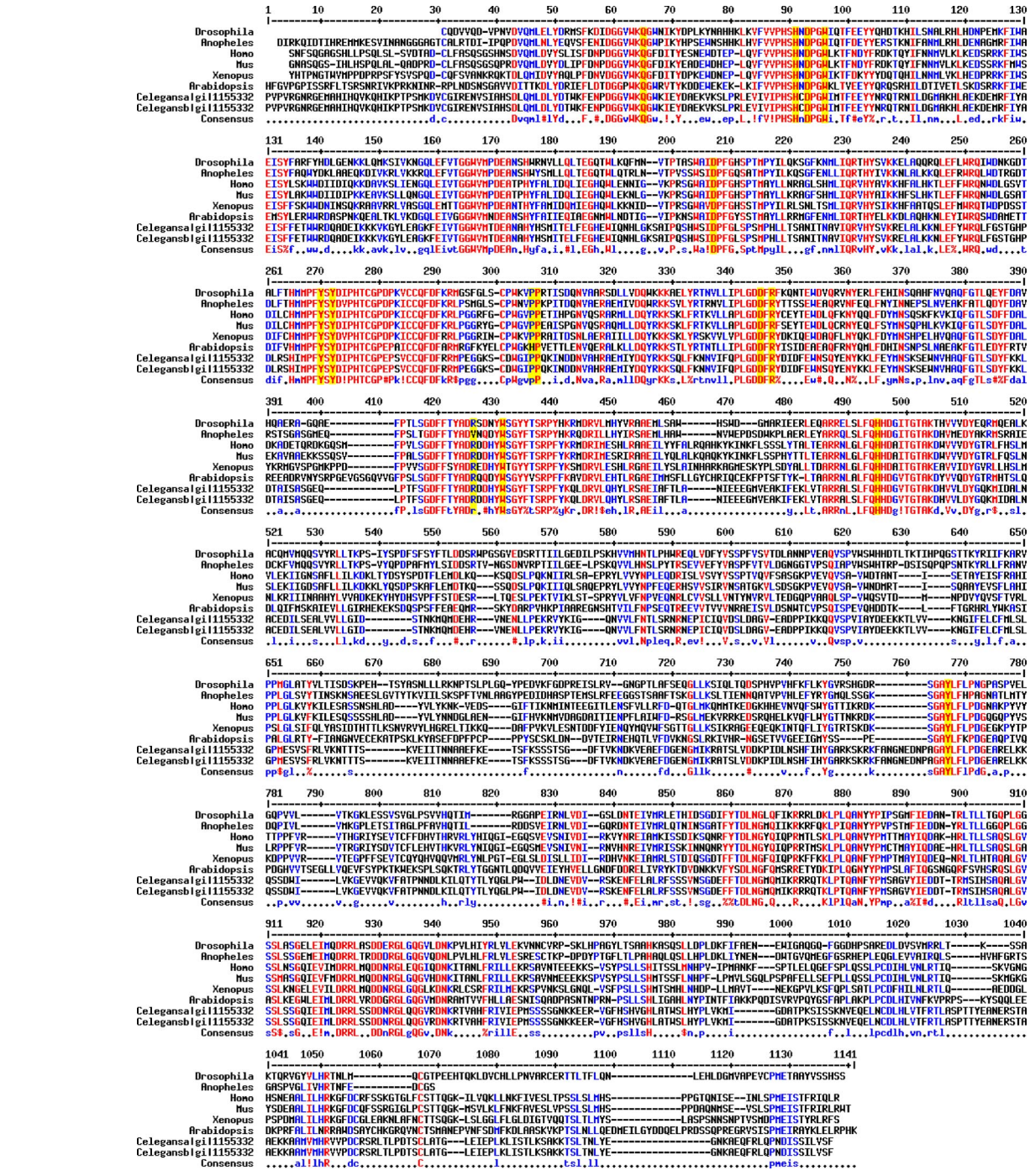

Fig. S1. A multiple sequence alignment comparing amino acid sequences of GMII from *Drosophila melanogaster*, *Anopheles gambia*, *Homo sapiens*, *Mus musculus*, *Xenopus laevis*, *Arabidopsis thaliana*, and *Caenorhabditis elegans*. Highlighted residues are essential for substrate binding and catalysis.

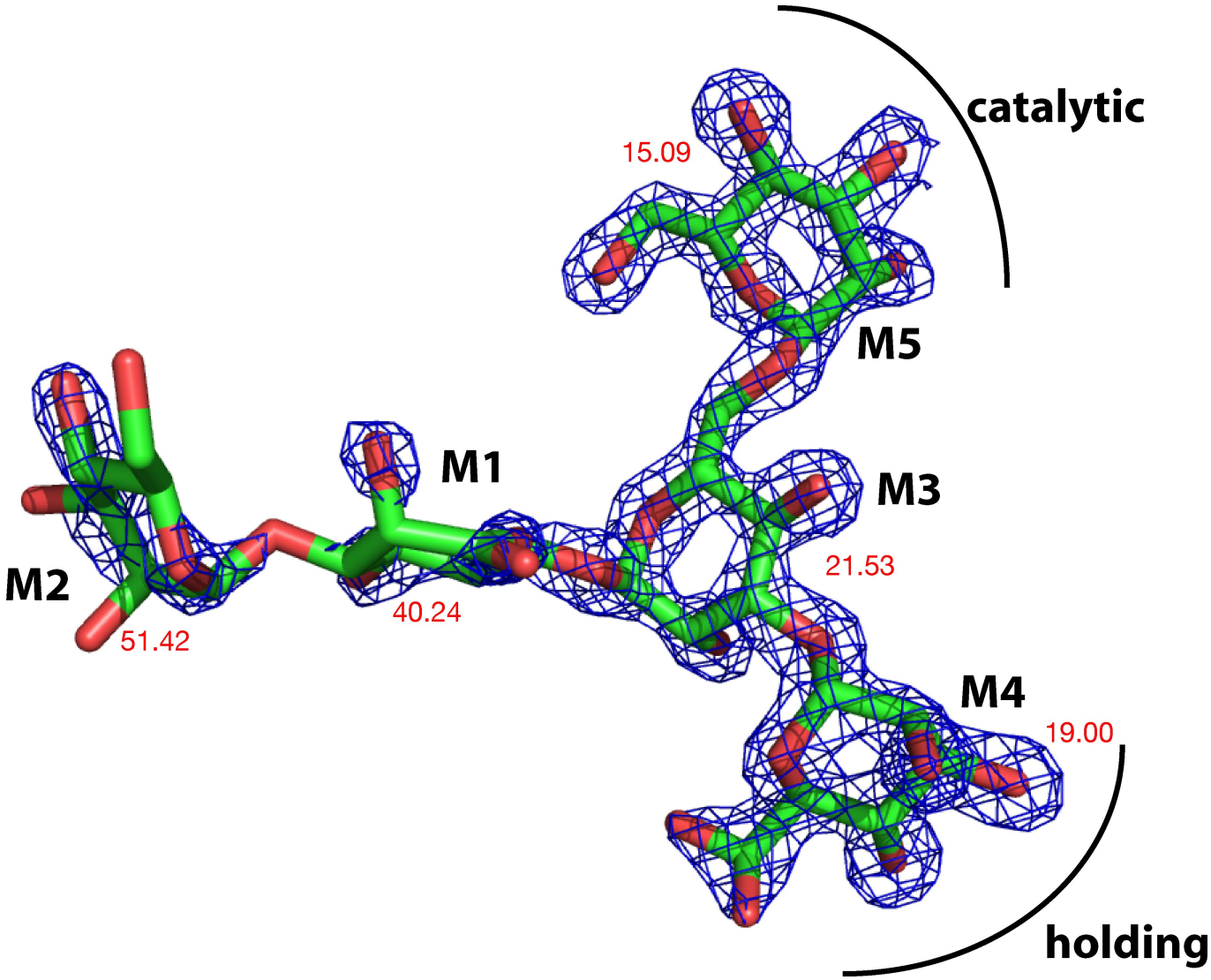

Fig. S2. Man_5 fitted to the $F_o - F_c$ simulated annealing omit map electron density in the dGMII active site. The electron density is contoured to 3.0 σ to show the quality of the density in the region of the catalytic and holding sites. When contoured at 2.0 σ, density for the complete saccharide is present.

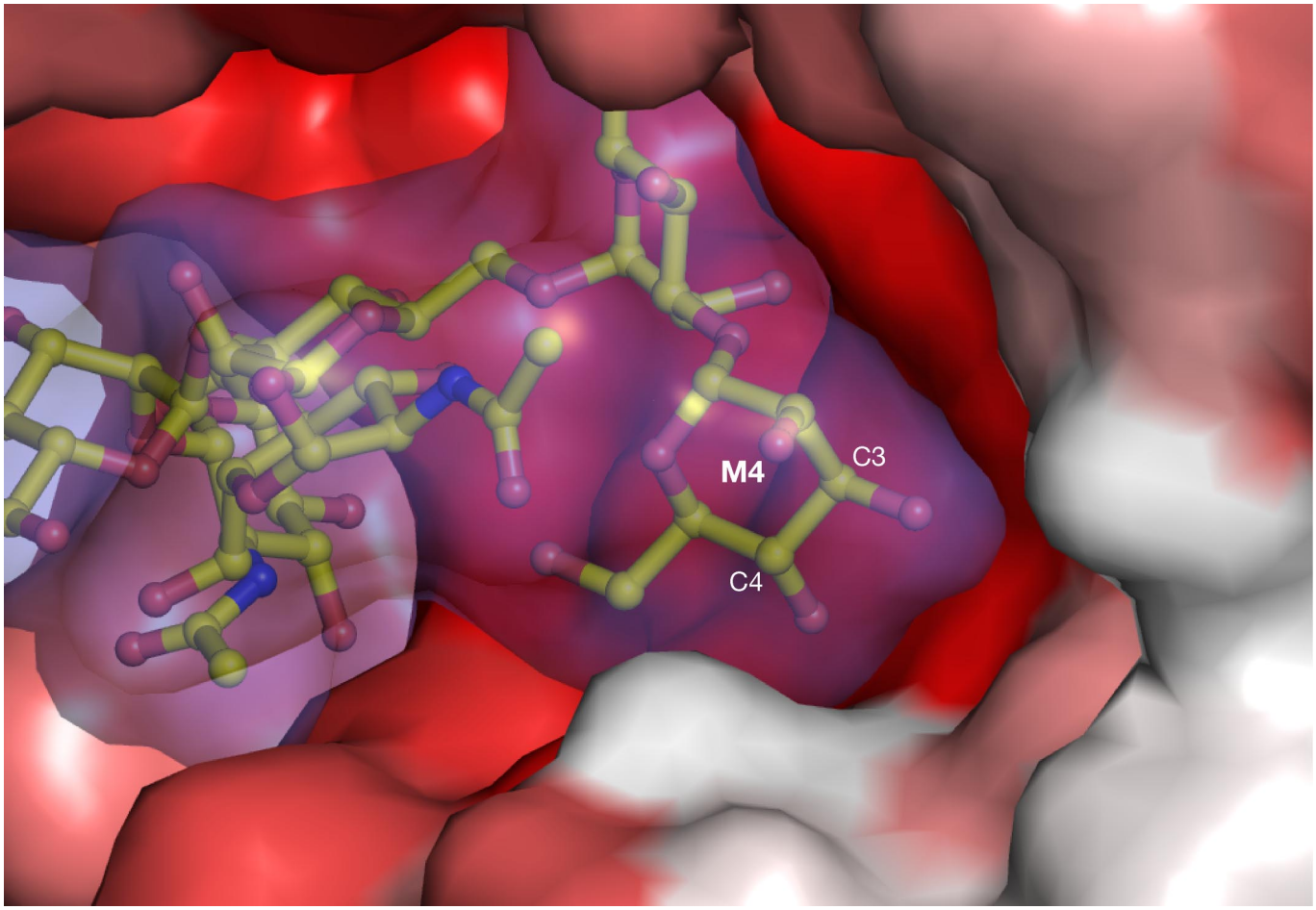

Fig. S3. Oligosaccharide position M4 (α1,3-linked mannose) packing tightly into the holding subsite. Positions 3 and 4 of M4 pack tightly against a wall of conserved residues, particularly R343 (not shown). The coloring of the protein reflects sequence conservation as determined by a multiple sequence alignment.

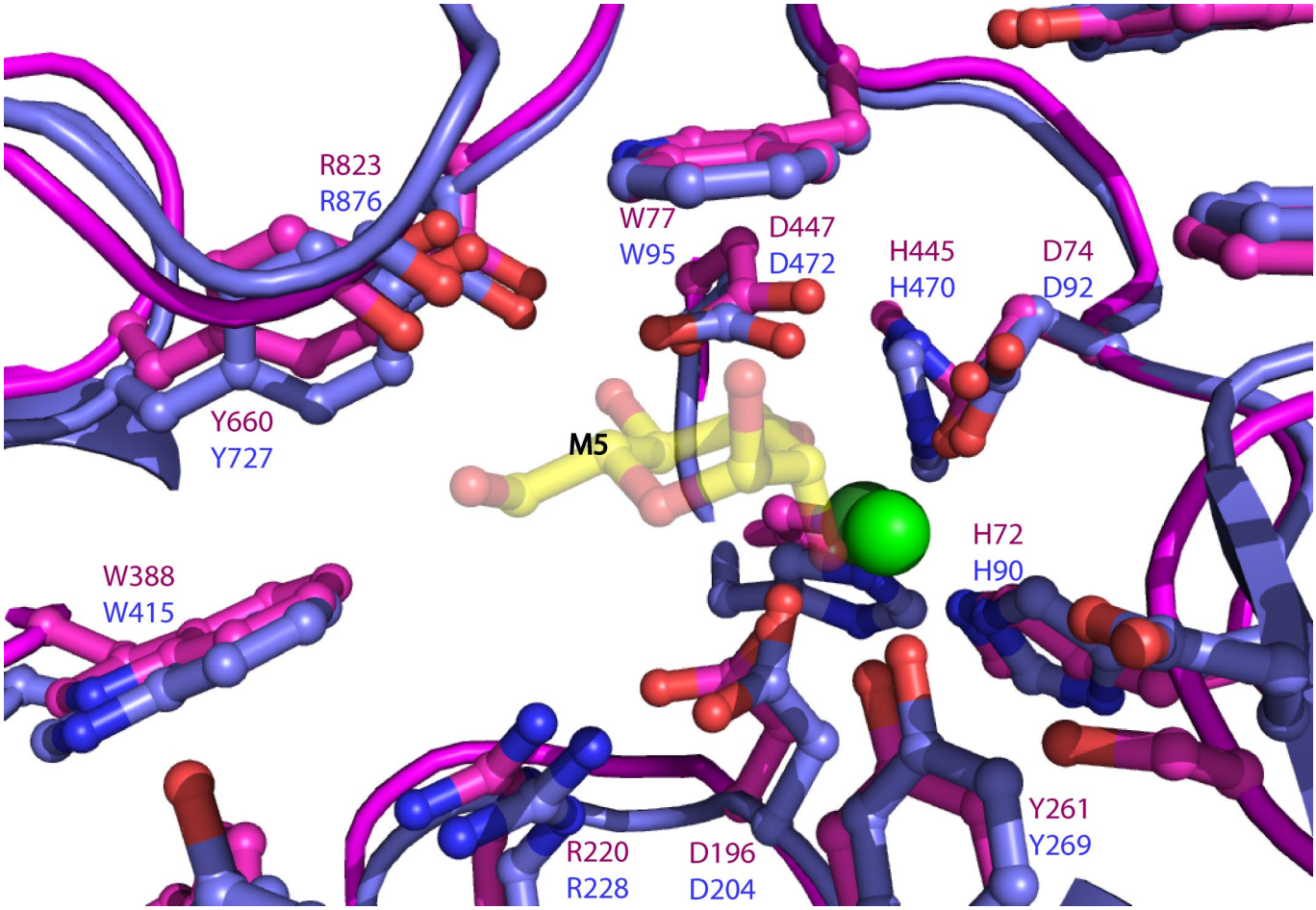

Fig. S4. A comparison of the atomic structures of the dGMII (blue) and bLM (magenta) catalytic sites. Relevant protein side chains are shown in stick representation and are numbered and colored respective to their protein. The M5 position as solved from the dGMII-substrate structure is presented in transparent yellow. Note that the side chains of the catalytic sites superimpose quite favorably, particularly those residues involved directly in saccharide coordination.

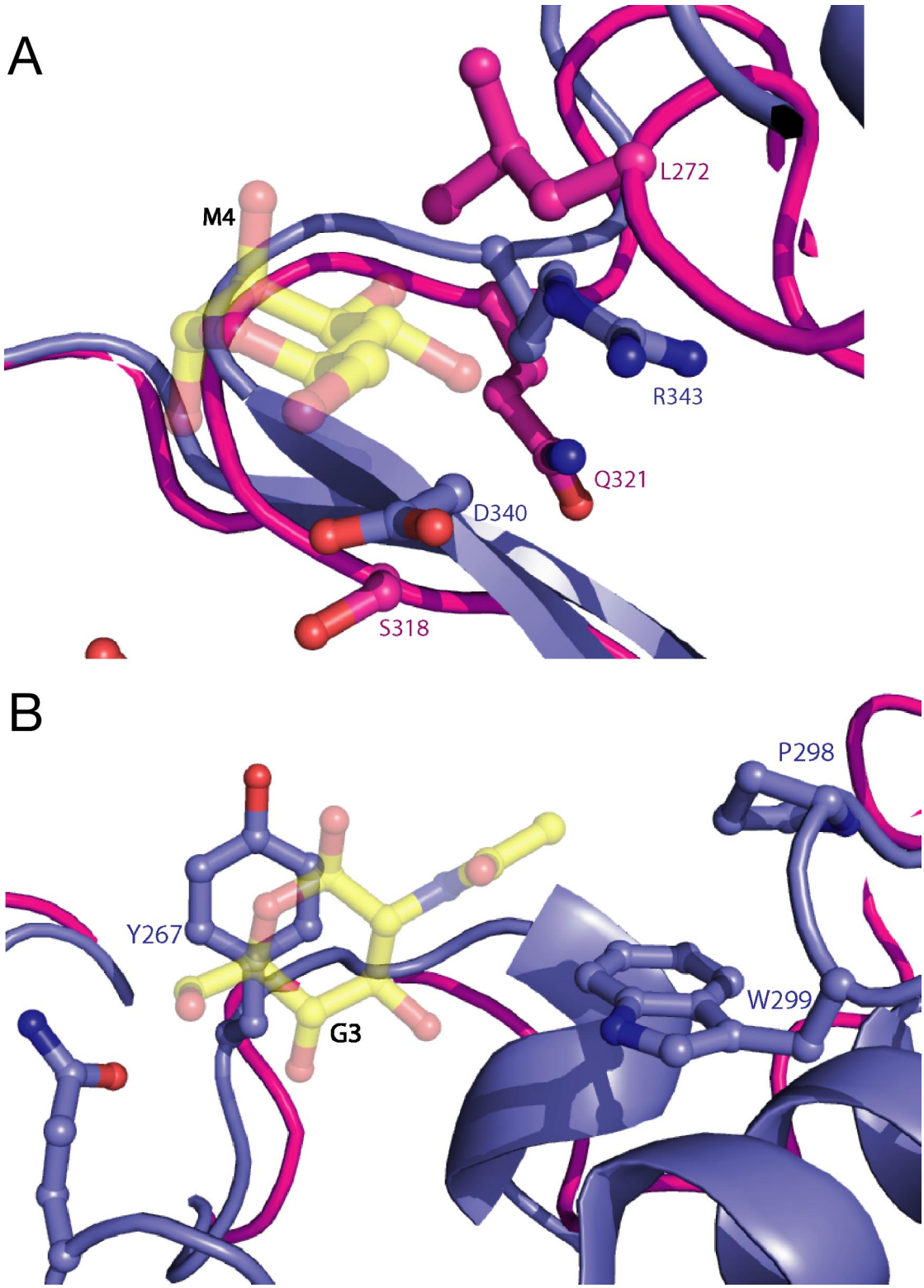

Fig. S5. Comparison of the atomic structures of the dGMII (blue) holding (*A*) and anchor (*B*) sites and the equivalent regions in bLM (magenta). Relevant protein side chains are shown in stick representation and are numbered and colored respective to their protein. The M4 and G3 positions as solved from the dGMII–substrate structure are presented in transparent yellow

Table S1. Data collection and refinement statistics

	dGMII(D204A)·GnMan$_5$Gn	dGMII(D204A)·Man$_5$	dGMII(D204A)
Data collection			
Space group	$P2_12_12_1$	$P2_12_12_1$	$P2_12_12_1$
Cell dimensions			
a, *b*, c, Å	68.9, 109.6, 139.0	68.7, 110.0, 139.3	68.8, 109.9, 138.5
α, β, γ, °	90, 90, 90	90, 90, 90	90, 90, 90
Resolution, Å	30–1.40 (1.44–1.40)	30–1.60 (1.62–1.60)	30–1.30 (1.39–1.30)
R_{sym}	0.10 (0.49)	0.07 (0.28)	0.11 (0.33)
$I/\sigma I$	14.7 (4.9)	16.5 (5.5)	11.2 (3.7)
Completeness, %	99.94 (99.55)	98.5 (96.5)	99.4 (96.9)
Redundancy	9.2 (7.0)	5.9 (5.0)	6.8 (4.0)
Refinement			
Resolution, Å	1.40	1.60	1.30
No. reflections	194,389	135,195	238,288
R_{work}/R_{free}	0.187 /0.209	0.159 /0.180	0.192 /0.216
No. atoms			
Protein	8434	8,375	8,415
Ligand	84	57	Not applicable
Water	1216	1,190	1,499
B factors			
Protein	15.51	15.35	11.25
Ligand	19.11	29.2	Not applicable
Water	29.51	28.3	28.92
rms deviations			
Bond lengths, Å	0.008	0.015	0.007
Bond angles, °	1.21	1.6	1.21
PDB structure			
ID	3CZN	3CVS	3CZS

Number in parentheses are for the highest-resolution shells.

Table S2. dGMII–GnMan$_5$Gn interactions

Oligosaccharide		Protein		
Residue	Atom	Residue	Atom	Interaction type
M5	O2	Y269	OH	Hydrogen bond
M5	O3	H471	NE2	Hydrogen bond
M5	O4	Y727	OH	Hydrogen bond
M5	O2	Zn		Metal coordination
M5	O3	Zn		Metal coordination
M5		W95		Stacking
M5		Y727		Stacking
M4	O3	R343	NE	Hydrogen bond
M4	O4	R343	NE	Hydrogen bond
M4	O4	R343	NH_2	Hydrogen bond
M4		D340		Stacking
M2		Q64		Stacking
M1	O4	Y267	OH	Hydrogen bond
G3	O3	H273	NE2	Hydrogen bond
G3		Y267		Stacking
G3		P298		Stacking
G3		P299		Stacking

Table S3. Key residues in the dGMII catalytic site with their structurally aligned bLM equivalents

dGMII	bLM	Role
W95	W77	M5 stacking
Y269	Y261	M5 H-bonding
H471	H446	M5 H-bonding
D472	D447	M5 H-bonding
Y727	Y660	M5 H-bonding
Zn	Zn	M5 coordination
H90	H72	Zn coordination
D92	D74	Zn coordination
H470	H445	Zn coordination
H471	H446	Zn coordination
D204	D196	Nucleophile