

Supplementary Information

Altered receptor specificity and cell tropism of D222G haemagglutinin mutants from fatal cases of pandemic A(H1N1) 2009 influenza

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Figure S1. Carbohydrate microarray analyses of the H1N1pdm viruses at HA titres of 500 and 100.

Figure S2. Comparisons of virus binding at 2000 HA titre to selected sialyl glycan probes in carbohydrate microarrays.

Figure S3. Carbohydrate microarray analyses of G155E, single mutant viruses (Ath and Lis) and of the G155E/D222G double mutant, Lvi, analysed at 2000 HA titres.

Table S1. Oligosaccharide probes included in the microarrays sorted by sialyl linkage and backbone sequence, and the binding signals (fluorescence intensities) they elicited with the six H1N1v viruses, Mol, Ham, Dak, Ham-e, Lvi and Nor.

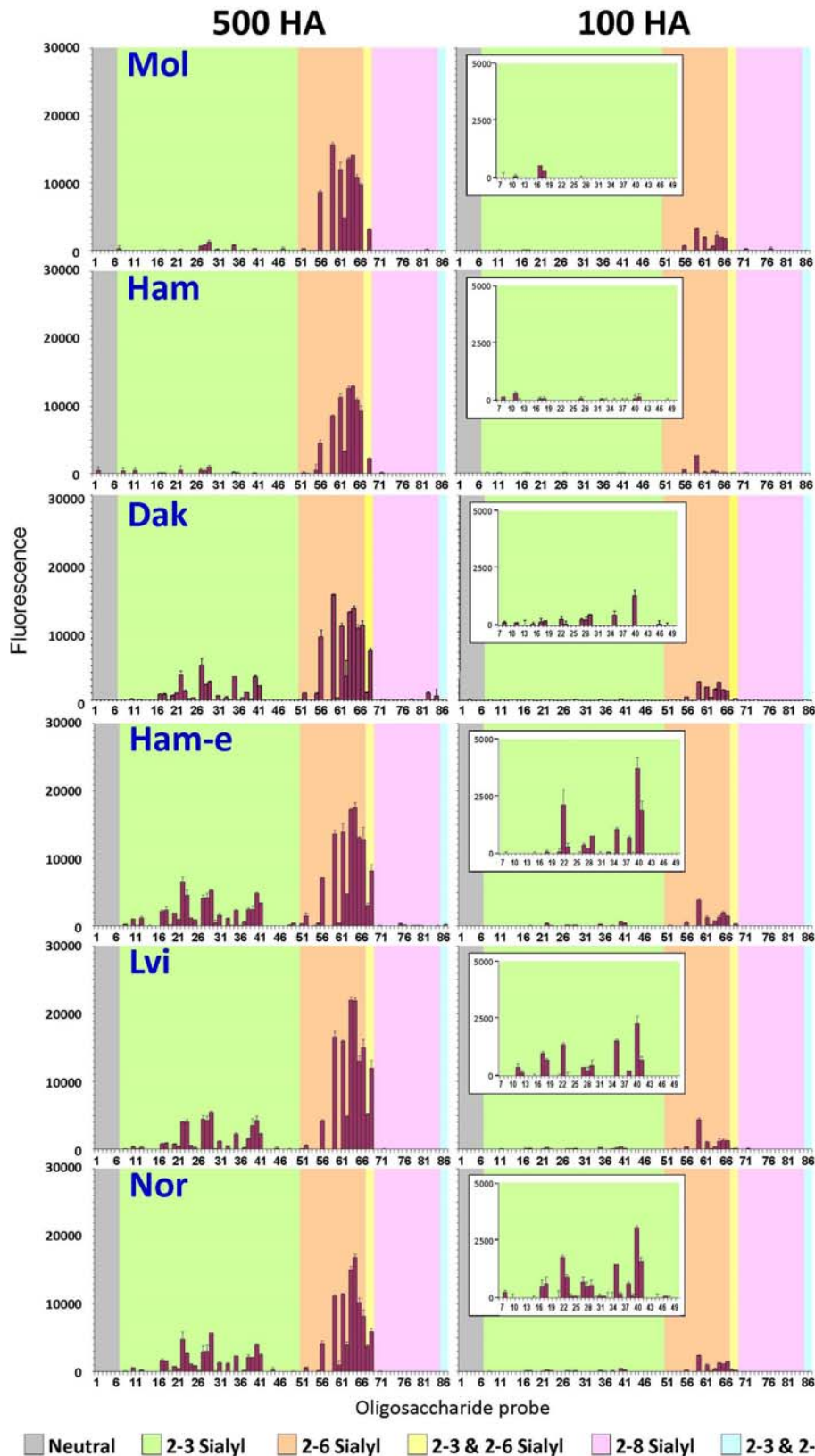


Figure S1. Carbohydrate microarray analyses of the H1N1pdm viruses at HA titres of 500 and 100. The two 222D (Mol and Ham), the 222E (Dak) and the three 222G (Ham-e, Lvi, and Nor) viruses were analysed also at HA titres of 500 and 100. In the 100 HA panels, the insets are fluorescence profiles at a higher (1.2 fold) laser power scan. Data for the 222D and 222G viruses are from experiments using the same virus preparations as in Fig 2, and for Dak from a different virus preparation.

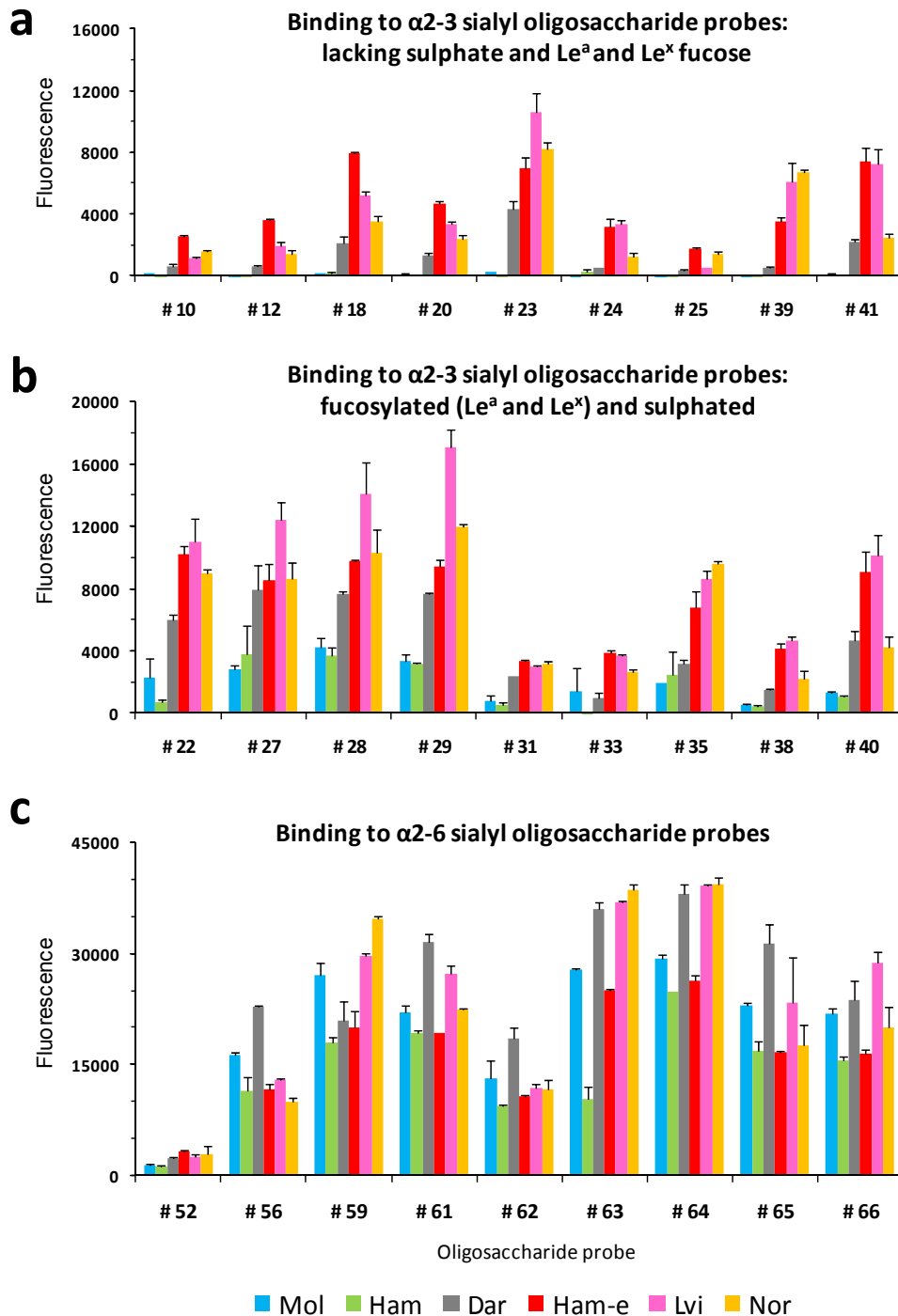


Figure S2. Comparisons of virus binding at 2000 HA to selected sialyl oligosaccharide probes in carbohydrate microarrays. Results are taken from Supplementary Table 1, and illustrate numerical scores for the binding signals (means of duplicate spots at 5 fmol/spot, with error bars) to compare the mutant with the 222D viruses. Colour codes for the viruses are as defined at the bottom of the figure. In panel **a** are results with non-sulphated α 2-3 sialyl oligosaccharide probes lacking the SLe^a and SLe^x sequences: the 222D viruses (Mol and Ham) show little or no binding to these; the 222E virus (Dak) shows weaker binding than do the 222G mutants (Ham-e, Lvi and Nor), and resembles the 222D viruses in showing negligible binding to probe 39 (VIM-2 antigen) known to be expressed on microvilli of the ciliated epithelium¹. In panel **b** are α 2-3 sialyl oligosaccharide probes having the SLe^a or SLe^x sequences with/without sulphation to which the 222D viruses bind weakly relative to the 222G mutants and Dak shows an intermediate binding strength. In panel **c** are selected α 2-6sialyl oligosaccharide probes showing similar binding profiles overall for the viruses investigated.

¹Loveless, R. W., Griffiths, S., Fryer, P. R., Blauth, C. & Feizi, T. Immunoelectron microscopic studies reveal differences in distribution of sialo-oligosaccharide receptors for *Mycoplasma pneumoniae* on the epithelium of human and hamster bronchi. *Infect. Immun.* **60**, 4015-4023 (1992).

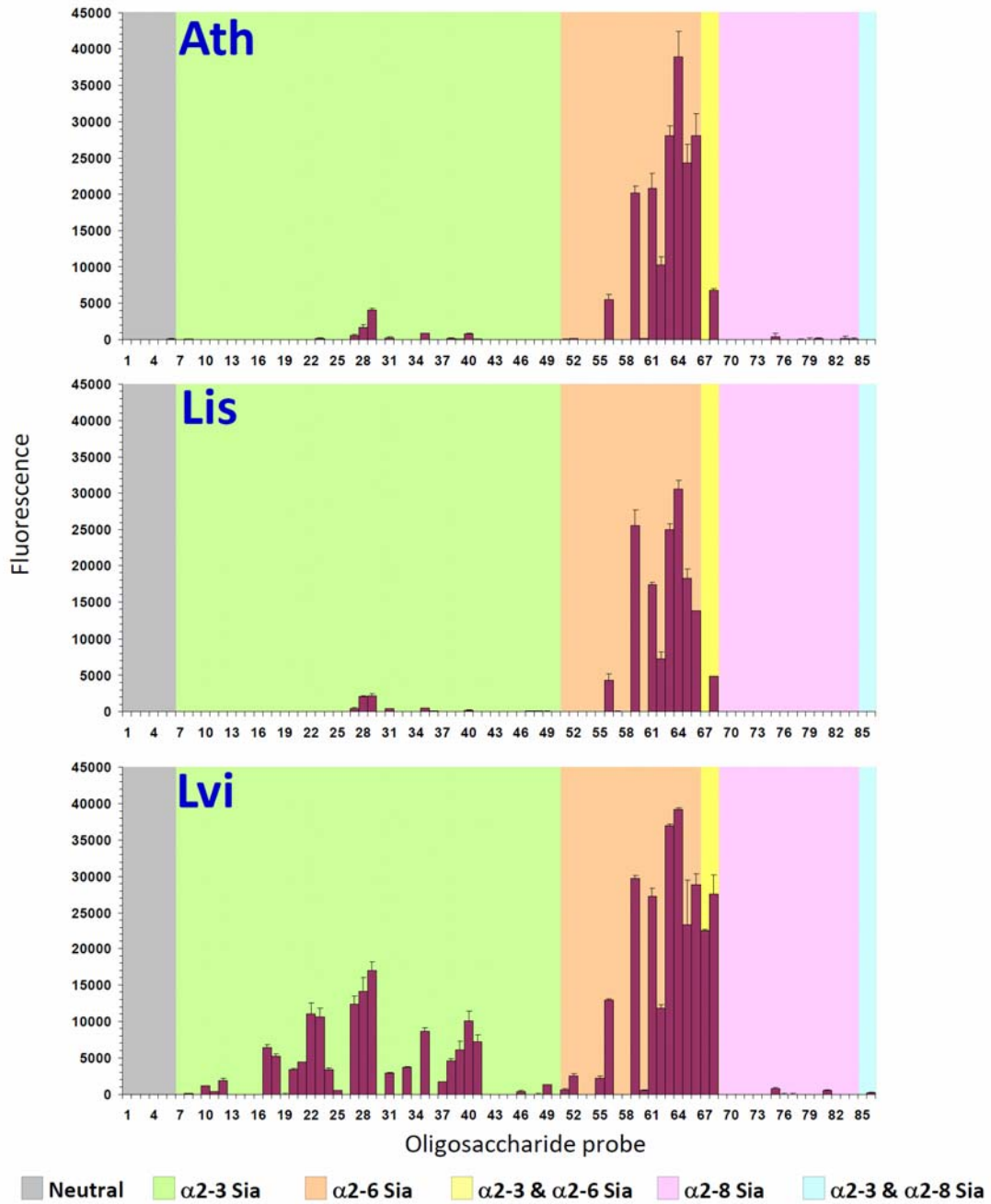


Figure S3. Carbohydrate microarray analyses of G155E, single mutant viruses (Ath and Lis) and of the G155E/D222G double mutant, Liv, analysed at 2000 HA titres.

Supplementary Table S1: Oligosaccharide probes included in the microarrays sorted by sialyl linkage and backbone sequence, and the binding signals (fluorescence intensities) they elicited with the six H1N1pdm viruses, Mol, Ham, Dak, Ham-e, Lvi, and Nor.

No.	Sequence ^a	Designation ^b	Fluorescence signal intensities at ~5 fmol ^c					
			Mol	Ham	Dak	Ham-e	Lvi	Nor
<i>Neutral</i>								
1	Galβ-4GlcNAc-DH	LacNAc	-	- ^d	-	-	-	-
2	Galβ-3GlcNAcβ-3Galβ-4Glc-DH	LNT	-	466	-	-	-	-
3	Galβ-4GlcNAcβ-3Galβ-4Glc-DH	LNnT	-	56	-	-	-	-
4	Galβ-4GlcNAcβ-3Galβ-4Glc-DH Fucα-3	LNFP-III	-	-	-	-	-	-
5	Galβ-4GlcNAcβ-6 Galβ-4Glc-DH Galβ-3GlcNAcβ-3	LNH	-	8	-	-	-	-
6	Galβ-4GlcNAcβ-2Manα-6 Manβ-4GlcNAcβ-4GlcNAc-DH Galβ-4GlcNAcβ-2Manα-3	NA2	-	-	-	-	-	-
<i>2,3-linked sialyl</i>								
Gal, Lactose and N-acetylactosamine-based linear backbones								
7	NeuAcα-3Galβ-Cer	GM4	-	-	-	98	-	-
			312	-	-	-	-	-
8	NeuAcα-3Galβ-4Glcβ-Cer	GM3	-	-	199	1,178	82	63
			-	420	-	208	89	60
			-	33	37	-	7	37
			-	137	120	-	-	208
9	NeuGcα-3Galβ-4Glc-Cer	GM3(Gc)	-	-	-	174	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-

10	NeuAc α -3Gal β -4Glc β -Cer	Haematoside	122	-	587	2,591	1,167	1,539
			-	-	164	970	391	578
			-	-	-	-	-	-
			-	-	-	-	-	21
11	NeuAc α -3Gal β -4Glc-DH	NeuAc α -(3')Lac	-	36	196	1,096	415	38
			-	485	-	77	107	-
			29	61	2	-	47	-
			111	291	118	-	390	-
12	NeuAc α -3Gal β -4Glc-AO	NeuAc α -(3')Lac-AO	-	-	640	3,636	1,952	1,369
			-	-	100	1,130	335	228
			-	20	-	-	19	-
			-	-	-	-	146	-
13	NeuAc β -3Gal β -4Glc-AO	NeuAc β -(3')Lac-AO	-	275	-	218	-	-
			-	-	-	25	-	-
			-	-	7	-	-	-
			-	-	-	-	3	-
14	Neu α -3Gal β -4Glc-AO	Neu α -(3')Lac-AO	-	-	9	263	-	-
			-	-	-	33	-	-
			-	-	23	-	-	-
			-	-	-	-	-	-
15	Neu4,5Ac α -3Gal β -4Glc-DH	Neu4,5Ac-(3')Lac	-	-	-	-	-	-
			-	-	-	-	-	-
			-	-	35	-	-	-
			-	-	44	-	-	-
16	Neu4,5Ac α -3Gal β -4Glc-AO	Neu4,5Ac-(3')Lac-AO	-	-	-	-	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
17	NeuAc α -3Gal β -3GlcNAc-DH	NeuAc α -(3')LN1-3	81	104	3,638	5,807	6,433	2,502
			31	151	792	2,124	759	1,650
			78	28	19	27	153	93
			502	87	117	-	1,005	485
18	NeuAc α -3Gal β -3GlcNAc-AO	NeuAc α -(3')LN1-3-AO	147	197	2,138	7,976	5,177	3,476
			35	31	769	2,361	886	1,610
			60	39	50	-	114	122
			273	70	178	103	654	645
19	NeuAc α -3Gal β -4GlcNAc-DH	NeuAc α -(3')LN	-	-	38	1,504	-	9
			-	-	-	148	-	20
			-	-	-	-	-	-
			-	-	-	-	-	-
20	NeuAc α -3Gal β -4GlcNAc-AO	NeuAc α -(3')LN-AO	106	81	1,266	4,641	3,361	2,347
			-	-	628	1,902	750	732
			-	-	-	6	-	-
			-	-	-	-	-	-

21	NeuAc α -3Gal β -3GlcNAc-AO Fuca-4	SA(3')-Lea-Tri-AO	730	337	1,790	5,857	4,371	3,307
			5	-	975	927	385	453
			-	-	4	15	14	2
			-	-	-	50	-	-
22	NeuAc α -3Gal β -4GlcNAc-AO Fuca-3	SA(3')-Lex-Tri-AO	2,249	673	5,966	10,224	10,989	9,013
			156	522	3,683	6,523	4,059	4,783
			-	-	56	385	273	285
			-	-	258	2,161	1,387	1,736
Lacto-N-neotetraose and Lacto-N-tetraose-based linear backbones								
23	NeuAc α -3Gal β -3GlcNAc β -3Gal β -4Glc-DH	LSTa	232	-	4,273	6,988	10,603	8,229
			16	-	1,236	4,595	4,057	2,756
			-	-	8	76	27	166
			-	-	23	336	34	947
24	NeuAc α -3Gal β -4GlcNAc β -3Gal β -4Glc β -Cer	Sialylparagloboside	-	212	543	3,194	3,376	1,222
			-	-	200	1,093	544	1,071
			-	-	-	-	-	10
			-	-	-	-	-	111
25	NeuAc α -3Gal β -4GlcNAc β -3Gal β -4Glc β -Cer36	GSC-31	-	-	369	1,782	485	1,407
			-	-	269	912	241	814
			-	-	-	-	-	5
			-	-	-	-	-	40
26	Neu α -3Gal β -4GlcNAc β -3Gal β -4Glc β -Cer36 SU-6	GSC-516B	-	-	-	-	-	-
			-	-	-	-	-	102
			-	-	-	-	-	-
			-	-	-	-	-	-
27	NeuAc α -3Gal β -4GlcNAc β -3Gal β -3GlcNAc-DH SU-6 SU-6 SU-6	C4U [Sialyl keratan sulphate]	2,838	3,816	7,921	8,524	12,385	8,641
			607	533	5,122	4,118	4,447	2,937
			-	44	42	119	85	133
			-	91	236	380	355	687
28	NeuAc α -3Gal β -3GlcNAc β -3Gal β -4Glc-DH Fuca-4	SA(3')-LNFP-II [SLe ^a]	4,235	3,651	7,661	9,768	14,112	10,264
			793	359	2,267	4,194	4,286	3,021
			-	-	50	56	75	101
			-	-	218	223	238	461
29	NeuAc α -3Gal β -4GlcNAc β -3Gal β -4Glc-DH Fuca-3	SA(3')-LNFP-III [SLe ^x]	3,376	3,120	7,650	9,436	17,041	11,970
			1,236	976	2,595	5,252	5,468	5,718
			-	-	73	151	107	112
			-	-	443	776	481	518
30	NeuAc α -3Gal β -4GlcNAc β -3Gal β -Cer36 Fuca-3	GSC-105	-	-	88	306	-	-
			-	-	-	571	39	66
			-	-	-	-	-	-
			-	-	-	-	-	-

31	$\begin{array}{c} \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-64 [SLe ^x (Cer)]	785	542	2,386	3,353	2,943	3,123
			159	-	611	1,574	1,139	1,286
			-	-	-	21	-	21
			-	-	-	-	-	104
32	$\begin{array}{c} \text{Neu}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-472	-	-	-	-	-	-
			-	-	-	-	-	-
			-	12	-	-	-	-
			-	49	-	-	-	63
33	$\begin{array}{c} \text{SU-6} \\ \\ \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-268 [6'Su-SLe ^x (Cer)]	1,394 ^e	-	917	3,873	3,679	2,616
			11	-	333	1,078	529	1,161
			-	-	-	-	-	-
			-	-	-	52	-	12
34	$\begin{array}{c} \text{SU-6} \\ \\ \text{Neu}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-268 deNAc	658	-	176	431	-	-
			-	-	60	94	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
35	$\begin{array}{c} \text{SU-6} \\ \\ \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-269 [6Su-SLe ^x (Cer)]	1,906	2,416	3,179	6,798	8,575	9,627
			740	207	3,423	2,318	2,220	2,269
			-	-	72	218	274	233
			-	-	399	1,051	1,551	1,438
36	$\begin{array}{c} \text{SU-6} \\ \\ \text{Neu}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-406	39	6	31	71	-	-
			31	57	-	83	29	-
			-	-	-	4	-	29
			-	-	-	-	-	189
37	$\begin{array}{c} \text{SU-6} \quad \text{SU-6} \\ \quad \\ \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-270	-	-	1,392	1,571	1,660	630
			29	-	309	648	268	195
			-	11	-	-	-	-
			-	-	-	-	-	-
38	$\begin{array}{c} \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \quad \\ \text{Fuca}\alpha\text{-3} \quad \text{Fuca}\alpha\text{-3} \end{array}$	GSC-220 [SLe ^x Le ^x (Cer)]	544	455	1,464	4,162	4,653	2,161
			17	-	1,078	2,448	1,544	2,153
			-	13	-	108	57	98
			-	34	-	674	207	626
39	$\begin{array}{c} \text{NeuAc}\alpha\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4GlcNAc}\beta\text{-3Gal}\beta\text{-4Glc}\beta\text{-Cer36} \\ \\ \text{Fuca}\alpha\text{-3} \end{array}$	GSC-221 [VIM-2]	-	-	492	3,507	6,094	6,669
			-	-	115	2,484	3,551	2,129
			-	-	-	19	129	4
			-	-	-	-	-	42
Branched backbone								
40	$\begin{array}{c} \text{Fuca}\alpha\text{-3} \\ \\ \text{Gal}\beta\text{-4GlcNAc}\beta\text{-6} \\ \\ \text{Gal}\beta\text{-4Glc-DH} \\ \\ \text{NeuAc}\alpha\text{-3Gal}\beta\text{-3GlcNAc}\beta\text{-3} \end{array}$	MSMFLNH	1,341	1,045	4,659	9,052	10,093	4,253
			282	102	3,367	4,805	4,303	3,951
			-	22	141	610	383	484
			-	101	1,278	3,721	2,298	3,090

N-glycans									
41	NeuAc α -3Gal β -4GlcNAc β -2Man α -6	A2F(2-3)	110	29	2,184	7,398	7,227	2,425	
			Fuc α -6	-	-	2,061	3,366	2,244	2,493
			Man β -4GlcNAc β -4GlcNAc-DH	-	27	-	363	129	266
	NeuAc α -3Gal β -4GlcNAc β -2Man α -3		-	135	-	1,939	712	1,641	
Ganglioside-based									
42	GalNAc β -4Gal β -4Glc β -Cer	GM2	-	-	-	238	-	-	
			-	-	-	-	-		
	NeuAc α -3		-	-	-	-	-		
43	NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc β -Cer	GM1b	-	-	-	-	-	-	
	-		-	-	-	-	-		
	-		-	-	-	-	-		
44	Gal β -3GalNAc β -4Gal β -4Glc β -Cer	GM1	-	-	-	-	-	-	
			-	-	-	-	295		
	NeuAc α -3		-	-	-	-	-		
45	Gal β -3GalNAc β -4Gal β -4Glc-DH	GM1-penta	-	-	-	-	-	-	
			-	-	-	144	-		
	NeuAc α -3		-	-	-	-	-		
46	Gal β -3GalNAc β -4Gal β -4Glc β -Cer	GM1(Gc)	-	-	-	-	359	-	
			-	-	7	-	-		
	NeuGc α -3		-	-	76	-	-		
47	Gal β -3GalNAc β -4Gal β -4Glc-DH	GM1(Gc)-penta	-	-	-	548	-	-	
			211	-	-	-	-		
	NeuGc α -3		-	-	-	-	-		
48	NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc β -Cer	GD1a	-	-	53	540	37	1	
			-	-	-	91	-		
	NeuAc α -3		-	-	-	-	-		
49	NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc-DH	GD1a-hexa	-	-	173	1,933	1,267	720	
			-	-	-	449	-		
	NeuAc α -3		-	-	-	-	-		
50	GalNAc β -4Gal β -3GalNAc β -4Gal β -4Glc β -Cer	GalNAc-GD1a(Ac,Gc)	-	-	-	-	-	-	
			-	-	-	-	-		
			NeuGc α -3	NeuAc α -3	-	-	-	-	
	GalNAc β -4Gal β -3GalNAc β -4Gal β -4Glc β -Cer		-	-	-	-	-		
	NeuAc α -3	NeuGc α -3	-	-	-	-			

2,6-linked sialyl

Gal, Lactose and N-acetylglucosamine-based linear backbones

51	NeuAc α -6Gal β -4Glc-DH	NeuAc α -(6')Lac	-	-	539	1,727	629	-
			-	-	51	273	48	-
			-	-	-	-	-	-
52	NeuAc α -6Gal β -4Glc-AO	NeuAc α -(6')Lac-AO	1,426	1,089	2,320	3,259	2,478	2,784
			284	159	998	1,482	575	625
			-	-	3	39	-	2
53	NeuAc β -6Gal β -4Glc-AO	NeuAc β -(6')Lac-AO	-	-	-	113	-	-
			-	35	-	-	-	-
			-	27	29	-	29	3
54	Neu α -6Gal β -4Glc-AO	Neu α -(6')Lac-AO	-	-	58	285	-	-
			-	-	-	15	-	-
			-	-	11	-	-	-
55	NeuAc α -6Gal β -4GlcNAc	NeuAc α -(6')LN	1,490	641	2,460	3,475	2,240	890
			-	508	953	406	-	93
			-	-	-	-	-	-
56	NeuAc α -6Gal β -4GlcNAc-AO	NeuAc α -(6')LN-AO	16,230	11,355	22,808	11,677	12,945	9,892
			8,657	4,536	9,213	7,146	4,227	4,170
			685	545	488	570	392	320
57	Neu5,9Ac α -6Gal β -4GlcNAc-DH	Neu5,9Ac-(6')LN	-	-	-	62	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-

Lacto-N-neotetraose and Lacto-N-tetraose-based linear backbones

58	Gal β -3GlcNAc β -3Gal β -4Glc-DH NeuAc α -6	LSTb	-	-	-	552	-	-
			-	-	-	3	-	-
			-	-	-	-	-	-
59	NeuAc α -6Gal β 4-GlcNAc β 3-Gal β 4-Glc-DH	LSTc	27,079	17,891	20,995	20,078	29,679	34,667
			15,667	8,620	15,416	13,724	16,623	11,166
			3,210	2,571	2,685	3,807	4,329	2,397
60	NeuAc α -6Gal β -4GlcNAc β -3Gal β -4Glc β -Cer36 Fu α -3	GSC-97	106	-	1,047	1,234	470	453
			15	-	304	481	-	970
			-	-	33	-	-	-
61	NeuAc α -6Gal β -4GlcNAc β -3Gal β -4Glc-DH Fu α -3	SA(6')-LNFP-VI	22,099	19,247	31,537	19,177	27,291	22,483
			12,011	11,384	10,819	13,930	15,938	11,512
			1,929	184	1,920	1,212	1,082	1,027

Branched backbones								
62	NeuAc α -6Gal β -4GlcNAc β -6	MSLNH	13,134	9,329	18,480	10,720	11,868	11,575
	Gal β -4Glc-DH		4,792	3,304	3,454	4,733	4,797	3,994
	Gal β -3GlcNAc β -3		184	61	364	127	57	75
63	Gal β -4GlcNAc β -6	MSLNnH-I	27,869	10,366	36,042	25,040	36,986	38,582
	Gal β -4Glc-DH		13,574	12,681	12,815	17,285	22,036	15,073
	NeuAc α -6Gal β -3GlcNAc β -3		581	356	1,509	758	365	404
64	NeuAc α -6Gal β -4GlcNAc β -6	DSLNNH	29,348	24,781	38,001	26,247	39,174	39,344
	Gal β -4Glc-DH		14,052	12,986	13,438	17,600	21,964	16,866
	NeuAc α -6Gal β -4GlcNAc β -3		2,293	188	2,589	1,294	1,179	1,206
65	Gal β -4GlcNAc β -6	MFMSLNnH	22,930	16,759	31,318	16,565	23,343	17,547
	Fuc α -3 Gal β -4Glc-DH		10,832	10,930	10,546	13,075	13,008	10,206
	NeuAc α -6Gal β -3GlcNAc β -3		1,931	28	1,501	1,993	1,305	1,131
N-glycans								
66	NeuAc α -6Gal β -4GlcNAc β -2Man α -6	A2(2-6)	21,817	15,559	23,778	16,526	28,819	20,059
	Man β -4GlcNAc β -4GlcNAc-DH		9,694	9,312	10,973	12,816	15,021	8,226
	NeuAc α -6Gal β -4GlcNAc β -2Man α -3		1,657	111	1,343	1,366	1,244	1,517
2,3:2,6-linked sialyl								
O-glycans								
67	NeuAc α -3Gal β -3GalNAc-DH	DST	237	167	3,072	4,538	22,545	13,719
	NeuAc α -6		-	-	1,046	3,085	5,116	3,773
			-	-	-	2	81	308
N-glycans								
68	NeuAc α -3Gal β -4GlcNAc β -2Man α -6	A3	14,374	7,521	17,671	14,367	27,519	21,970
	Man β -4GlcNAc β -4GlcNAc-DH		3,080	2,244	7,205	8,206	11,918	5,881
	NeuAc α -3Gal β -4GlcNAc β -4Man α -3		25	65	229	298	160	219
	NeuAc α -6Gal β -4GlcNAc β -2							

2,8-linked sialyl Ganglioside-based								
69	NeuAc α -8NeuAc α -3Gal β -4Glc β -Cer	GD3	-	-	-	373	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
70	NeuAc α -8NeuAc α -3Gal β -4Glc-DH	GD3-tetra	-	-	-	460	-	-
			-	-	-	57	-	-
			-	-	-	-	-	-
71	NeuAc α -8NeuAc α -3Gal β -4Glc-AO	GD3-tetra-AO	64	148	553	436	-	-
			-	144	76	-	29	-
			207	77	58	-	105	-
72	GalNAc β -4Gal β -4Glc β -Cer NeuAc α -8NeuAc α -3	GD2	-	-	-	-	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
73	Gal β -3GalNAc β -4Gal β -4Glc β -Cer NeuAc α -8NeuAc α -3	GD1b	-	-	-	-	-	18
			-	-	-	-	-	-
			-	-	7	15	-	-
74	NeuAc α -8NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc β -Cer NeuAc α -8NeuAc α -3	GQ1b	13	-	-	334	-	59
			-	-	-	18	-	-
			-	-	-	-	14	-
Polysialyl								
75	NeuAc α -8NeuAc-DH	SA2(α 8)	81	-	413	2,217	797	802
			-	-	-	324	18	29
			-	11	-	-	-	-
76	NeuAc α -8NeuAc α -8NeuAc-DH	SA3(α 8)	-	-	206	1,125	-	383
			-	-	-	126	-	-
			-	4	-	-	-	-
77	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH	SA4(α 8)	-	77	-	503	33	-
			-	-	-	-	-	-
			333	-	-	-	-	-
78	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH ^f	SA5(α 8)	-	-	-	355	-	-
			-	-	45	48	-	-
			-	4	-	-	-	-
79	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH ^f	SA6(α 8)	-	-	-	1,186	-	-
			-	-	-	16	-	-
			-	33	-	-	-	-

80	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH ^f	SA7(α 8)	-	-	-	837	33	-
			-	-	-	95	-	-
			-	27	-	-	-	-
81	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-8NeuAc α -8NeuAc-DH ^f	SA8(α 8)	-	-	-	634	452	-
			-	-	-	-	-	-
			-	-	-	-	-	-
82	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-8NeuAc α -8NeuAc-8NeuAc-DH ^f	SA9(α 8)	68	-	964	142	-	-
			-	-	-	-	-	-
			-	-	-	-	-	-
83	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH ^f	SA10(α 8)	-	-	49	976	-	34
			-	-	-	-	-	-
			-	-	-	-	-	-
84	NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc α -8NeuAc-DH ^f	SA11(α 8)	-	-	-	956	-	-
			-	-	561	36	-	-
			-	-	-	-	-	-
2,3:2,8-linked sialyl								
Ganglioside-based								
85	NeuAc α -8NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc β -Cer NeuAc α -3	GT1a	-	-	-	180	-	-
			-	-	-	-	-	-
			-	10	13	-	-	-
86	NeuAc α -3Gal β -3GalNAc β -4Gal β -4Glc β -Cer NeuAc α -8NeuAc α -3	GT1b	-	-	-	598	172	50
			-	-	-	185	-	-
			-	8	-	-	-	-

^a The oligosaccharide probes are all lipid-linked, and are from the collection assembled in the course of research in Glycosciences Laboratory. DH, designates NGLs prepared from reducing oligosaccharides by reductive amination with the amino lipid, 1,2-dihexadecyl-*sn*-glycero-3-phosphoethanolamine (DHPE)¹; AO, NGLs prepared from reducing oligosaccharides by oxime ligation with an aminoxy-functionalized DHPE²; Cer, natural glycolipids with various ceramide moieties; Cer36, synthetic glycolipids with ceramide having a total of 36 carbon atoms. NB, Neu denotes de-N-acetylated neuraminic acid.

^b Alternative designations for probes described in Table 2 are in brackets.

^c For each probe fluorescence signal intensities are shown for 2000, 500 and 100 HA titres. In the shaded rows, fluorescence signals at 100 HA are shown at higher laser power scan (as in Fig. S2 insets) for the α 2-3-linked sialyl probes (positions 7-50).

^d -, Signal less than 1.

^e Artefact on the slide at the position of probe 33 in the experiment of Mol at 2000 HA. This value is taken as 'less than 500' in Table 2 to avoid confusion.

^f Major component.

1. Chai, W., Stoll, M. S., Galustian, C., Lawson, A. M. & Feizi, T. Neoglycolipid technology - deciphering information content of glycome. *Methods Enzymol.* **362**, 160-195 (2003).
2. Liu, Y. *et al.* Neoglycolipid probes prepared via oxime ligation for microarray analysis of oligosaccharide-protein interactions. *Chem. Biol.* **14**, 847-859 (2007).