

Table 1. O-Glycans found in leukemia cell lines

Structure and monosaccharide compositions	Molecular ions	Peak ID	Relative abundance (%)				
			K562	U937	Jurkat HL-60		
Asialo glycan							
Gal β 1-3(Gal β 1-4GlcNAc β 1-6)GalNAc-2AA#A2	869(+)	H-1			4.44		
Monosialo glycans							
NeuAc α 2-3Gal β 1-3GalNAc-2AA#MS1 (Sialyl T antigen)	795(+)	K-1, J-1, H-4	U1,	32.3	36.6	20.5	15.2
NeuAc α 2-6GalNAc-2AA (Sialyl Tn antigen)	633(+)	H-7, J-2			28.9	1.16	
NeuAc α 2-3Gal β 1-3(GlcNAc β 1-6)GalNAc-2AA#MS2	997(+)	H-3				4.24	
Gal β 1-3(Gal β 1-4GlcNAc β 1-6)GalNAc-2AA + NeuAc $_1$ #MS3	1159(-)	H-6				4.26	
NeuAc α 2-3Gal β 1-3(GlcNAc β 1-6)GalNAc-2AA + Fuc	1143(-)	H-2				2.08	
Gal β 1-3(Gal-(Fuc-)GlcNAc β 1-6)GalNAc-2AA + NeuAc $_1$ #MS4	1305(-)	H-5				2.26	
Disialo glycans							
NeuAc α 2-3Gal β 1-3(NeuAc α 2-6)GalNAc-2AA#DS1 (Disialyl T antigen)	1085(-)	K-3, J-5, H-14	U-3,	30.7	30.1	12.9	2.97
NeuAc α 2-3Gal β 1-3(NeuAc α 2-3Gal β 1-4GlcNAc β 1-6)GalNAc-2AA#DS2	1450(-)	J-4, H-13			10.7	25.9	
NeuAc α 2-3Gal β 1-3(NeuAc α 2-3Gal β 1-4(Fuca1-3)GlcNAc β 1-6)GalNAc-2AA	1596(-)	H-12				0.14	
NeuAc α 2-3Gal β 1-3(NeuAc α 2-3Gal β 1-4GlcNAc β 1-3Gal β 1-4GlcNAc β 1-6)GalNAc-2AA#DS3	1815(-)	H-10				1.49	
NeuAc α 2-3Gal β 1-3(NeuAc α 2-3Gal β 1-4GlcNAc β 1-3Gal β 1-4(Fuca1-3)GlcNAc β 1-6)GalNAc-2AA#DS4	1961(-)	H-10				1.49	
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) $_2$ -Gal β 1-4GlcNAc β 1-6)GalNAc-2AA#DS6	2180(-)	H-9				1.11	
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) $_3$ -Gal β 1-4GlcNAc β 1-6)GalNAc-2AA#DS9	2545(-)	H-8				2.13	
Degradation product							
NeuAc α 2-3Gal-2AA	592(+)	K-2, J-3, H-11	U-2,	17.3	21.2	20.6	23.7

We showed ms/ms data for the glycan with # markes in supporting information. The information is available free of charge via the Internet at <http://pubs.acs.org>. The structures in blue are confirmed according to the analogous consideration on the structures of higher / lower series of O-glycans. The structures in red are not assigned in the present study, because we could not observe good MSⁿ data.

Table 2. O-Glycans found in pancreatic cancer cell lines

O-glycans observed in PANC1	Molecular ions	O-glycans observed in BxPC3	Molecular ions
<u>Asialo fraction(peak P-1): 0%</u>		<u>Asialo fraction (Peak B-1):6.0%</u> Galβ1-3GalNAc-2AA Galβ1-3(GlcNAcβ1-6)GalNAc-2AA ^{#A1} Galβ1-3(Galβ1-4GlcNAcβ1-6)GalNAc-2AA ^{#A2} Galβ1-3(GlcNAcβ1-3Galβ1-4GlcNAcβ1-6)GalNAc-2AA ^{#A4} Galβ1-3(Gal-GlcNAc-Galβ1-4GlcNAcβ1-6)GalNAc-2AA ^{#A5}	504 (+) 707 (+) 869 (+) 1093(+) 1255(+)
<u>Monosialo fraction (Peak P2, P3): 63%</u> NeuAcα2-3Galβ1-3GalNAc-2AA ^{#MS1} (Sialyl T antigen)	795(+)	<u>Monosialo fraction (Peak B-2): 70%</u> NeuAcα2-3Galβ1-3GalNAc-2AA ^{#MS1} (Sialyl T antigen) Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS8} Galβ1-3(Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS5} Galβ1-3 (Gal-GlcNAc-Gal-(Fuc-) GlcNAcβ1-6) ·GalNAc-2AA + NeuAc ₁ ^{#MS6} Galβ1-3((Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS8} Galβ1-3((Gal-GlcNAc) ₂ -Gal-(Fuc-)GlcNAcβ1-6) ·GalNA-2AA + NeuAc ₁ ^{#MS9} Galβ1-3((Gal-GlcNAc) ₃ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS10} Galβ1-3((Gal-GlcNAc) ₃ -Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA + NeuAc ₁ ^{#MS11} Galβ1-3((Gal-GlcNAc) ₄ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS12} Galβ1-3((Gal-GlcNAc) ₄ -Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#MS13}	795(+) 1159(-) 1524(-) 1670(-) 1889(-) 2035(-) 2254(-) 2400(-) 2619(-) 2765(-)
<u>Disialo fraction (Peak P4): 37%</u> NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA ^{#DS1} (Disialyl T antigen)	1085(-)	<u>Disialo fraction (Peak B3, B4, B5): 24%</u> NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA ^{#DS1} (Disialyl T antigen) NeuAc₂Hex₁HexNAc₁Hex₁-2AA NeuAc-Galβ1-3(NeuAc-Gal-GlcNAcβ1-6)GalNAc-2AA ^{#DS2} NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc	1085(-) 1231(-) 1450(-) 1815(-)

-2AA#DS3		
NeuAc-Gal β 1-3(NeuAc-Gal-GlcNAc-Gal-(Fuc-)GlcNAc β 1-6)		1961(-)
GalNAc-2AA#DS4		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-GlcNAc β 1-6)		2180(-)
GalNAc-2AA#DS6		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-(Fuc-)GlcNAc β 1-6)		2326(-)
GalNAc-2AA#DS7		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₃ -Gal-GlcNAc β 1-6)		2545(-)
GalNAc-2AA#DS9		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₃ -Gal-(Fuc-)GlcNAc β 1-6)		2691(-)
GalNAc-2AA		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₄ -Gal-GlcNAc β 1-6)GalNAc		2910(-)
-2AA#DS10		
NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₄ -Gal-(Fuc-)GlcNAc β 1-6)		3056(-)
GalNAc-2AA		
<u>Degradation product</u>		
NeuAc α 2-3Gal-2AA	592(+)	592(+)

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Table 3. O-Glycans found in colon cancer cell lines

O-glycans observed in LS174T	Molecular ions	O-glycans observed in HCT-15	Molecular ions
<u>Asialo fraction(peak L-1): 25.6%</u>			
Galβ1-3GalNAc-2AA	504 (+)	GlcNAc-GalNAc-2AA	545 (+)
GlcNAc-GalNAc-2AA	545 (+)	Fuc-Galβ1-3GalNAc-2AA	649(+)
Galβ1-3(GlcNAcβ1-6)GalNAc-2AA#A1	707 (+)	Galβ1-3(GlcNAcβ1-6)GalNAc-2AA#A1	707 (+)
Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA#A2	869 (+)	Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA#A2	869 (+)
Galβ1-3(Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA#A3	1014(-)		
Galβ1-3(GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA#A4	1071(-)		
Galβ1-3(Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA#A5	1233(-)		
Galβ1-3(Gal-GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA#A6	1379(-)		
Galβ1-3(GlcNAc-Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA#A7	1436(-)		
Galβ1-3((Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA#A8	1598(-)		
Galβ1-3(((Gal-GlcNAc) ₂ -Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA#A9	1744(-)		
<u>Monosialo fraction (Peak L-2): 37.1%</u>			
Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS3	1159(-)	<u>Monosialo fraction (Peak H-2): 23.8%</u>	
Galβ1-3(Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS4	1305(-)	NeuAcc2-3Galβ1-3GalNAc-2AA#MS1 (Sialyl T antigen)	795(+)
Galβ1-3(Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1 + SO ₃	1385(-)	Galβ1-3(GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS2	997(-)
Galβ1-3(Gal-GlcNAc-Gal-(Fuc-)(SO ₃ -)GlcNAcβ1-6)GalNAc-2AA#SU2	1459(-)	Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS3	1159(-)
		Galβ1-3(Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS4	+ 1305(-)
Galβ1-3(Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS5	1524(-)		
Galβ1-3(Gal-(Fuc-)GlcNAc-Gal-(Fuc-)(SO ₃ -)GlcNAcβ1-6)GalNAc-2AA#SU8	1604(-)		
Galβ1-3(Gal-GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS6	1670(-)		
Galβ1-3(Gal-(Fuc-)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS7	1816(-)		
Galβ1-3(Gal-(Fuc-)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc1 + SO ₃	1896(-)		
Galβ1-3((Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1 + SO ₃	1969(-)		
Galβ1-3((Gal-GlcNAc) ₂ -Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA	2035(-)		

+ NeuAc ₁ ^{#MS9}			
Galβ1-3(Gal-GlcNAc-Gal-(Fuc-)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA + NeuAc ₁	2181(-)		
<u>Disialo fraction (Peak L-3, L-4, L-5): 37.1%</u>			
NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA ^{#DS1} (Disialyl T antigen)	1085(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAcβ1-6)GalNAc-2AA ^{#DS2}	1450(-)		
NeuAc-Galβ1-3(NeuAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA	1596(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-GlcNAcβ1-6) GalNAc-2AA ^{#DS3}	1815(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-(SO ₃ -)GlcNAcβ1-6) GalNAc-2AA ^{#SU4}	1895(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA ^{#DS4}	1961(-)		
NeuAc-Galβ1-3(NeuAc-Gal-(Fuc-)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA ^{#DS5}	2107(-)		
Galβ1-3((Gal-GlcNAc) ₂ -Gal-(Fuc-)(SO ₃ -)GlcNAcβ1-6) GalNA-2AA + NeuAc ₁ ^{#SU5}	2115(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA ^{#DS6}	2180(-)		
Galβ1-3(Gal-GlcNAc-Gal-(Fuc-)(SO ₃ -)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA + NeuAc ₁ ^{#SU6}	2260(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA ^{#DS7}	2326(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-(Fuc-)GlcNAc-Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA ^{#DS8}	2472(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₃ -Gal-(Fuc-)GlcNAcβ1-6) GalNAc-2AA	2691(-)		
<u>Degradation product</u>			
(Gal-GlcNAc) ₂ -Gal-2AA ^{#DP2}	1029(-)		
Gal-GlcNAc-Gal-(Fuc-)GlcNAc-Gal-2AA ^{#DP3}	1176(-)		
(Gal-GlcNAc) ₃ -Gal-2AA + Fuc	1541(-)		
NeuAc-Gal-(Fuc-)GlcNAc-Gal-(Fuc-)(SO ₃ -)GlcNAc-Gal-2AA ^{#DP6}	1691(-)		
<u>Disialo fraction (Peak H-3, H-4): 62.4%</u>			
NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA ^{#DS1} (Disialyl T antigen)	1085(-)		
Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + SO ₃ + NeuAc ₁ ^{#SU1}	1238(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAcβ1-6)GalNAc-2AA ^{#DS}	1450(-)		
Galβ1-3(Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA + SO ₃ + NeuAc ₁	1604		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-GlcNAcβ1-6) GalNAc-2AA ^{#DSS}	1815(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6) GalNAc-2AA ^{#DS6}	2180(-)		
2			
<u>Degradation product</u>			
NeuAc-Gal-GlcNAc-Gal-2AA ^{#DPI}	956(+)		

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Table 4. O-Glycans found in gastric cancer cell lines

O-glycans observed in MKN45	Molecular ions	O-glycans observed in MKN7	Molecular ions
<u>Asialo fraction (Peak M-1): 5.0%</u>			
Galβ1-3GalNAc-2AA	504 (+)		
GlcNAc-GalNAc-2AA	545 (+)		
Galβ1-3(GlcNAcβ1-6)GalNAc-2AA#A1	707 (+)		
<u>Monosialo fraction (Peak M-2, M-3): 21.3%</u>			
NeuAcα2-6GalNAc-2AA + Na (Sialyl Tn antigen)	654(+)	NeuAcα2-6GalNAc-2AA + Na (Sialyl Tn antigen)	654(+)
NeuAcα2-3Galβ1-3GalNAc-2AA#MS1 (Sialyl T antigen)	795(+)	NeuAcα2-3Galβ1-3GalNAc-2AA#MS1 (Sialyl T antigen)	795(+)
Galβ1-3(GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS2	998(+)	Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS3	1159(-)
Galβ1-3(Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS3	1159(-)	Galβ1-3(Gal-(Fuc-)GlcNAcβ1-6)GalNAc-2AA#MS4	1305
Galβ1-3(Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS5	1524(-)		
Galβ1-3((Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS8	1889(-)		
Galβ1-3((Gal-GlcNAc) ₃ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS10	2254(-)		
Galβ1-3((Gal-GlcNAc) ₄ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS12	2619(-)		
Galβ1-3((Gal-GlcNAc) ₅ -Gal-GlcNAcβ1-6)GalNAc-2AA + NeuAc1#MS14	2984(-)		
<u>Disialo fractions (Peak M-4, M-6): 61.4%</u>			
NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA#DS1 (Disialyl T antigen)	1085(-)	NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA#DS1 (Disialyl T antigen)	1085(-)
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAcβ1-6)GalNAc-2AA#DS2	1450(-)		
NeuAc-Galβ1-3(NeuAc-Gal-GlcNAc-Gal-GlcNAcβ1-6)GalNAc-2AA#DS3	1815(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₂ -Gal-GlcNAcβ1-6)GalNAc-2AA#DS6	2180(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₃ -Gal-GlcNAcβ1-6)GalNAc-2AA#DS9	2545(-)		
NeuAc-Galβ1-3(NeuAc-(Gal-GlcNAc) ₄ -Gal-GlcNAcβ1-6)GalNAc-2AA#DS10	2910(-)		
<u>Disialo fractions (Peak M7-5): 25.4%</u>			
		NeuAcα2-3Galβ1-3(NeuAcα2-6)GalNAc-2AA#DS1 (Disialyl T antigen)	1085(-)

NeuAc-Gal β 1-3(NeuAc-(Gal-GlcNAc) ₅ -Gal-GlcNAc β 1-6)GalINAc-2AA #DS11	3275(-)
<u>Trisialo fraction (Peak M-5): 12.3%</u>	
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA#TS1	2106(-)
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA+Gal-GlcNAc#TS2	2471(-)
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA+(Gal-GlcNAc) ₂ #TS3	2836(-)
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA+(Gal-GlcNAc) ₃ #TS4	3201(-)
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA+(Gal-GlcNAc) ₄ #TS5	3566(-)
NeuAc-Gal-GlcNAc-(NeuAc-Gal-GlcNAc)Galb1-3(NeuAca2-6)GalNAc-2AA+(Gal-GlcNAc) ₅ #TS6	3931(-)
<u>Degradation product</u>	
NeuAca2-3Gal-2AA	592(+)
NeuAc-Gal-GlcNAc-Gal-2AA#DP1	957(+)
NeuAc-(Gal-GlcNAc) ₂ -Gal-2AA#DP4	1321(-)
NeuAc-(Gal-GlcNAc) ₃ -Gal-2AA#DP5	1686(-)
NeuAc-(Gal-GlcNAc) ₄ -Gal-2AA#DP7	2051(-)
NeuAc-(Gal-GlcNAc) ₅ -Gal-2AA	2416(-)
NeuAc-(Gal-GlcNAc) ₆ -Gal-2AA#DP8	2781(-)
<u>Degradation product</u>	
NeuAca2-3Gal-2AA	592(+)
NeuAc-Gal-GlcNAc-Gal-2AA#DP1	956(+)

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Table 5 Mucin-type O-glycans observed in HCT116 cell.

M1		M4	
504	Gal-GalNAc-2AA	1815	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA
546	GlcNAc-GalNAc-2AA	1969	Gal β 1-3 {(Gal-GlcNAc) $_3\beta$ 1-6}GalNAc-2AA + NeuAc + SO₃
625	GlcNAc-GalNAc-2AA + SO₃	2180	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_3\beta$ 1-6}GalNAc-2AA
706	Gal β 1-3(GlcNAc β 1-6)GalNAc-2AA	2335	Gal β 1-3 {(Gal-GlcNAc) $_4\beta$ 1-6}GalNAc-2AA + NeuAc + SO₃
786	Gal β 1-3(GlcNAc β 1-6)GalNAc-2AA + SO₃	2546	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_4\beta$ 1-6}GalNAc-2AA
868	Gal β 1-3 (Gal-GlcNAc β 1-6)GalNAc-2AA	M5	
949	Gal β 1-3 (Gal-GlcNAc β 1-6)GalNAc-2AA + SO₃	1450	NeuAc α 2-3Gal β 1-3 (NeuAc α 2-6Gal-GlcNAc β 1-6)GalNAc-2AA
1234	Gal β 1-3 {(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA	1815	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA
M2		M6	
1158	Gal β 1-3 (Gal-GlcNAc β 1-6)GalNAc-2AA + NeuAc	1085	NeuAc α 2-3Gal β 1-3(NeuAc α 2-6)GalNAc-2AA
1361	Gal β 1-3 (GlcNAc-Gal-GlcNAc β 1-6)GalNAc-2AA + NeuAc	1238	Gal β 1-3 (Gal-GlcNAc β 1-6)GalNAc-2AA + NeuAc + SO₃
1523	Gal β 1-3 {(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA + NeuAc	1440	Gal β 1-3 (GlcNAc-Gal-GlcNAc β 1-6)GalNAc-2AA + NeuAc + SO₃
1888	Gal β 1-3 {(Gal-GlcNAc) $_3\beta$ 1-6}GalNAc-2AA + NeuAc	1450	NeuAc α 2-3Gal β 1-3 (NeuAc α 2-6Gal-GlcNAc β 1-6)GalNAc-2AA
2253	Gal β 1-3 {(Gal-GlcNAc) $_4\beta$ 1-6}GalNAc-2AA + NeuAc	1604	Gal β 1-3 {(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA + NeuAc + SO₃
2618	Gal β 1-3 {(Gal-GlcNAc) $_5\beta$ 1-6}GalNAc-2AA + NeuAc	1684	Gal β 1-3 {(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA + NeuAc + 2SO₃
M3		1764	Gal β 1-3 {(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA + NeuAc + 3SO₃
633	NeuAc α 2-6GalNAc-2AA	2260	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_3\beta$ 1-6}GalNAc-2AA + SO₃
795	NeuAc α 2-3Gal β 1-3GalNAc-2AA	2626	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_4\beta$ 1-6}GalNAc-2AA + SO₃
998	NeuAc α 2-3Gal β 1-3(GlcNAc β 1-6)GalNAc-2AA	2106	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_2\beta$ 1-6}GalNAc-2AA + NeuAc
1160	Gal β 1-3 (Gal-GlcNAc β 1-6)GalNAc-2AA + NeuAc	2471	NeuAc α 2-3Gal β 1-3 { NeuAc α 2-6(Gal-GlcNAc) $_3\beta$ 1-6}GalNAc-2AA + NeuAc

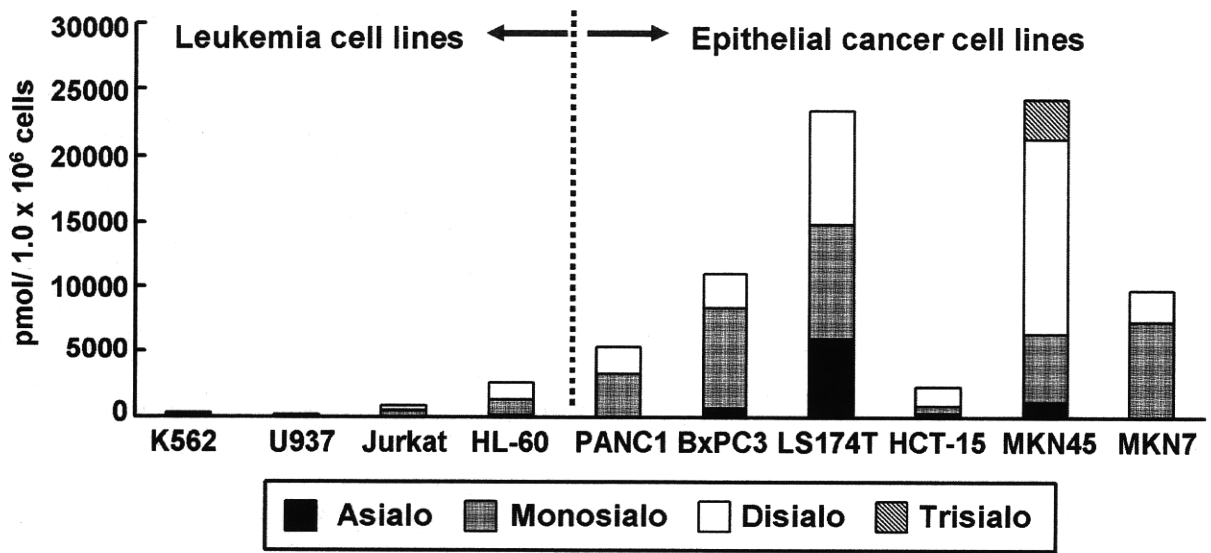


Fig.1 Comparison of the amounts of O-glycans expressed on cancer cells

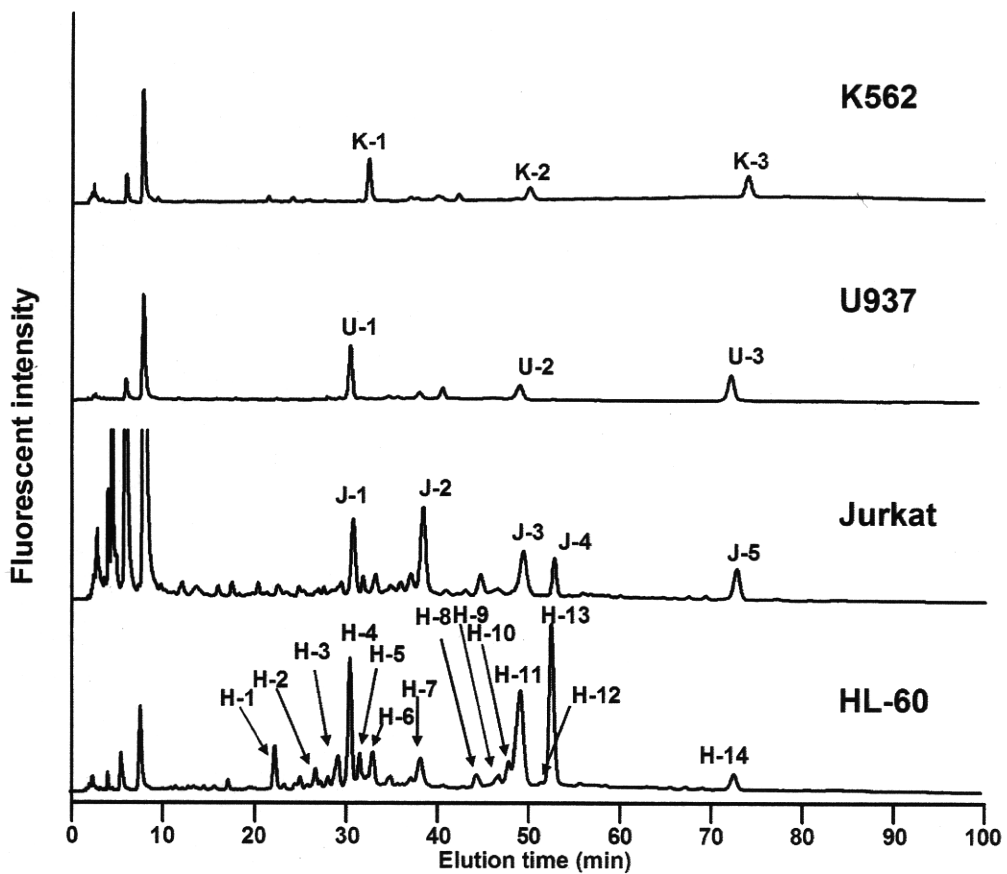


Fig.2 NP-HPLC analysis of O-glycans derived from leukemia cells

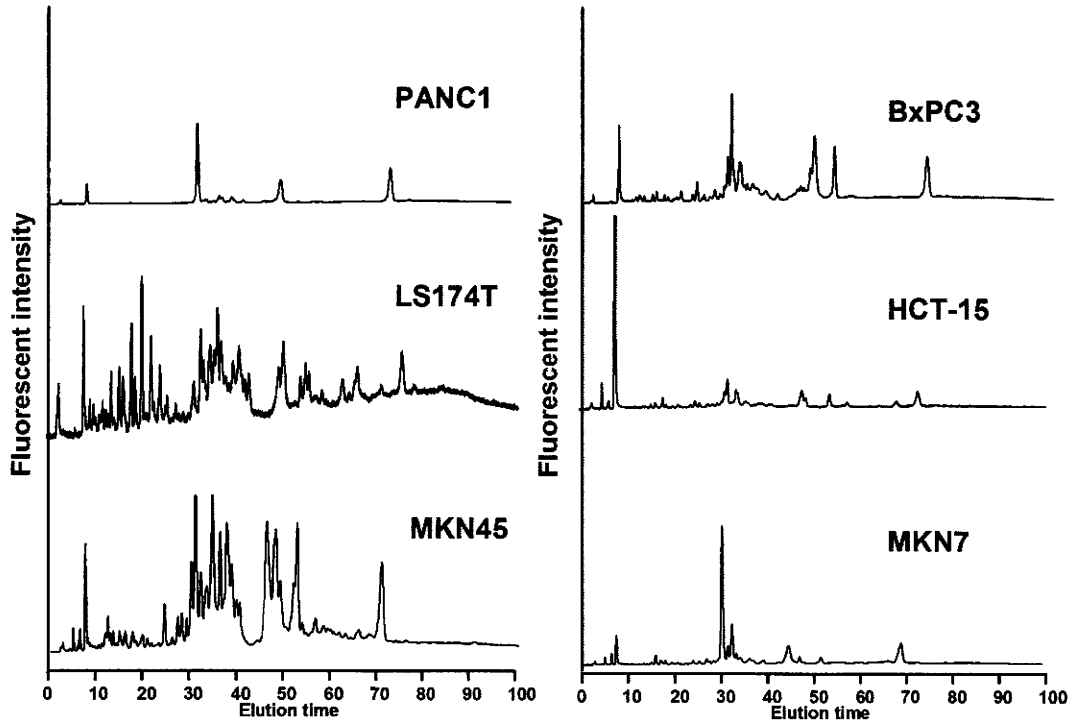


Fig.3 NP-HPLC analysis of O-glycans derived from epithelial cancer cells

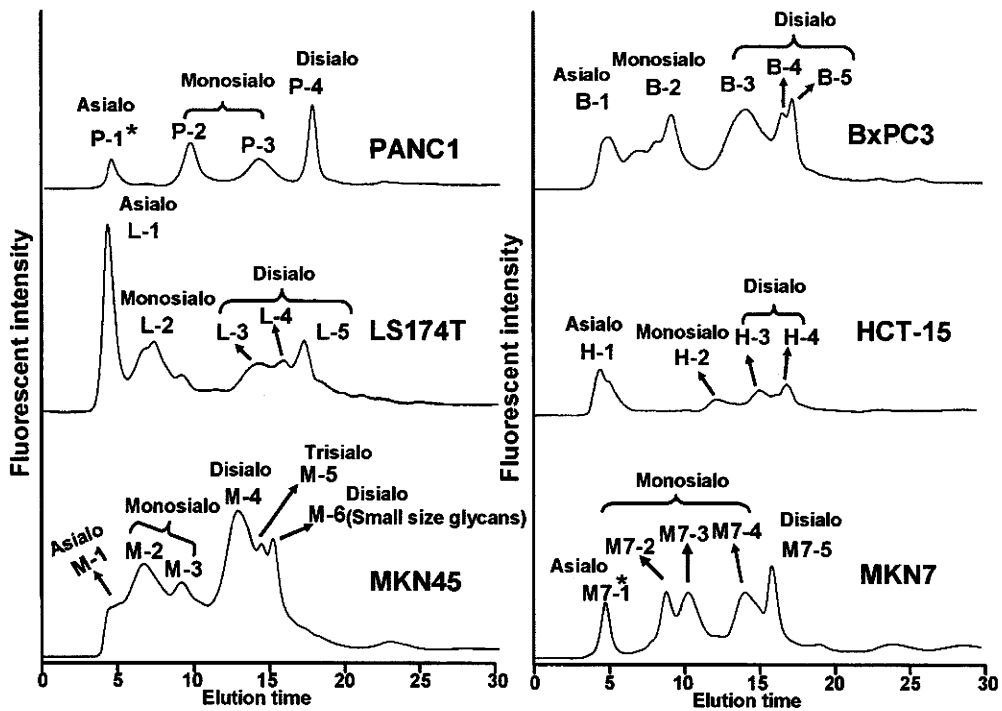


Fig.4 Separation of O-glycan pool derived from epithelial cancer cells by serotonin affinity chromatography

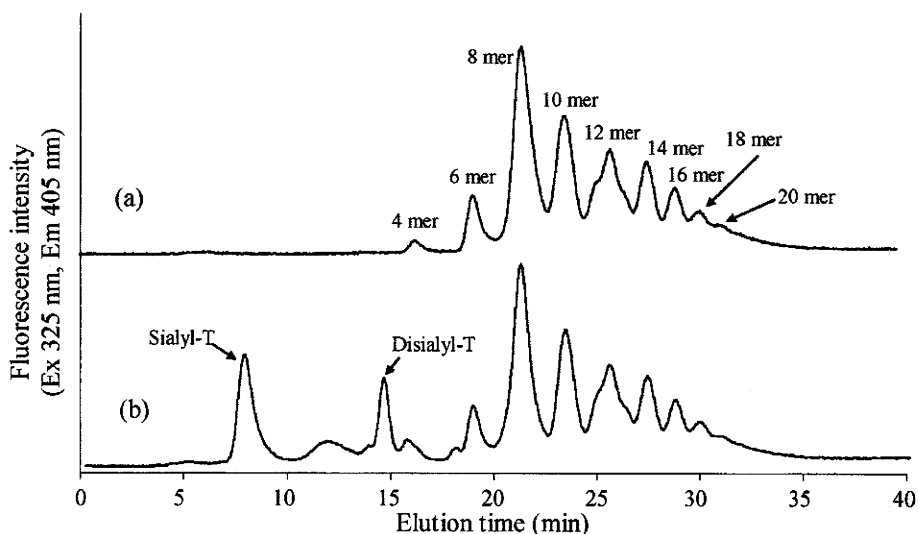


Fig.5 Serotonin affinity chromatography of HA oligosaccharides and mucin-type O-glycans. (a) 2AA-labeled HA oligosaccharides (4-20 mer) and (b) a mixture of mucin-type O-glycans from bovine fetuin and HA oligosaccharides. Analytical conditions; column, LA-serotonin (4.6 x 150 mm). flow rate, 0.5 mL/min. eluent; solvent A, water. solvent B, 50 mM Ammonium acetate in water. gradient conditions, a linear gradient (5-75 % solvent B) from 2 to 45 min and 75% solvent B from 37 to 45 min.

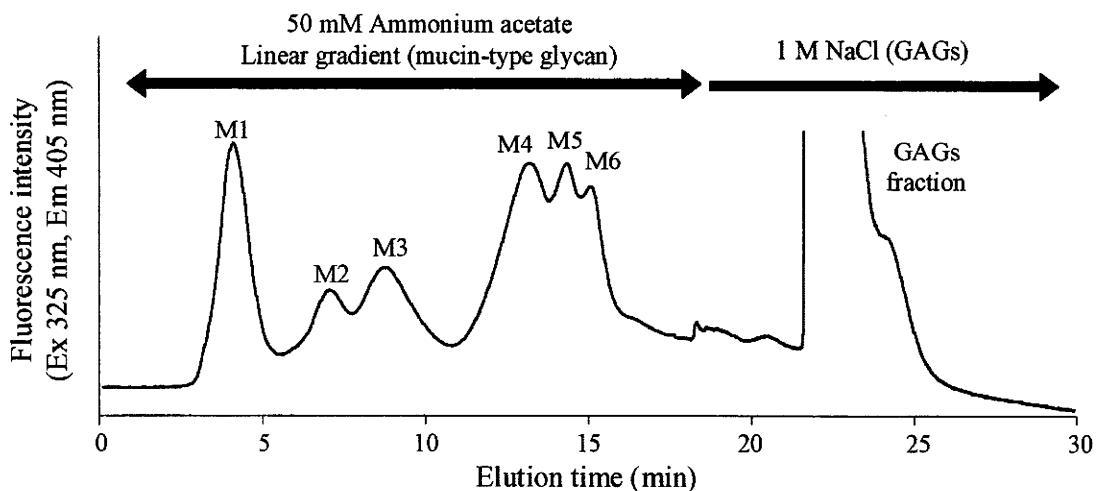


Fig.6 Serotonin affinity chromatography of O-linked glycans derived from HCT116 cell. Analytical conditions; column, LA-serotonin (4.6 x 150 mm). flow rate, 0.5 mL/min. eluent; solvent A, water. solvent B, 50 mM Ammonium acetate in water. gradient conditions, a linear gradient (5-40 % solvent B) from 2 to 20 min and 1 M NaCl from 20 to 45 min.

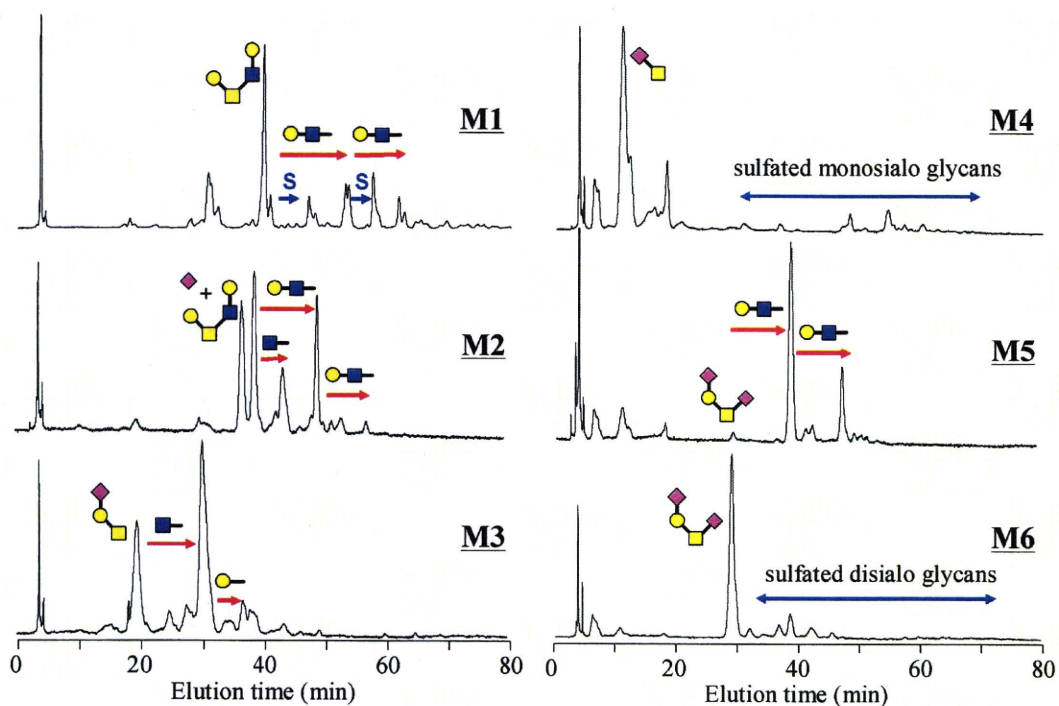


Fig.7 NP-HPLC analysis of mucin-type O-glycans from HCT116 cell. Analytical conditions: column, TSK-GEL Amide-80 (4.6 x 250 mm). flow rate, 0.8 mL/min. eluent, solvent A, 0.1% CH₃COOH in MeCN. solvent B, 0.2% CH₃COOH/0.2% triethylamine in water. gradient conditions: a linear gradient (15-50 % solvent B) from 5 to 85 min.

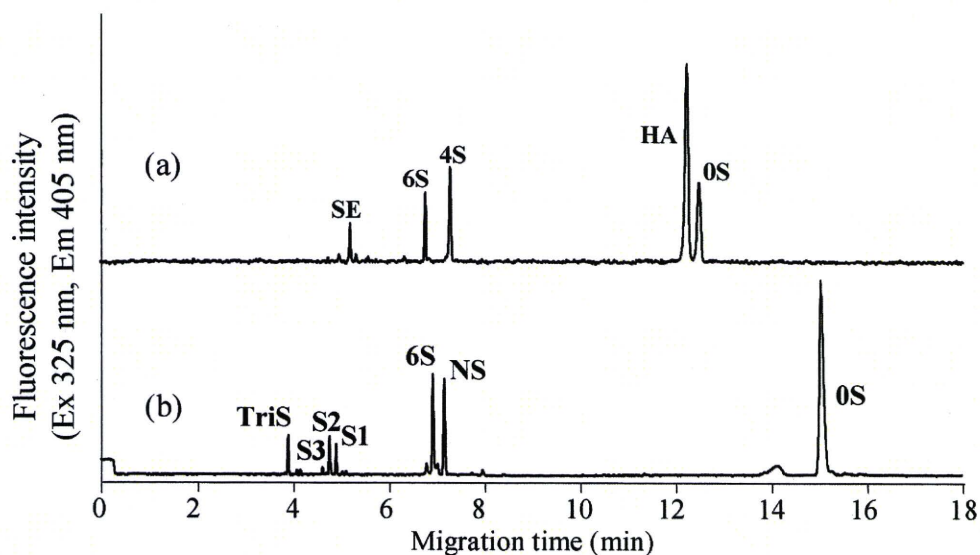
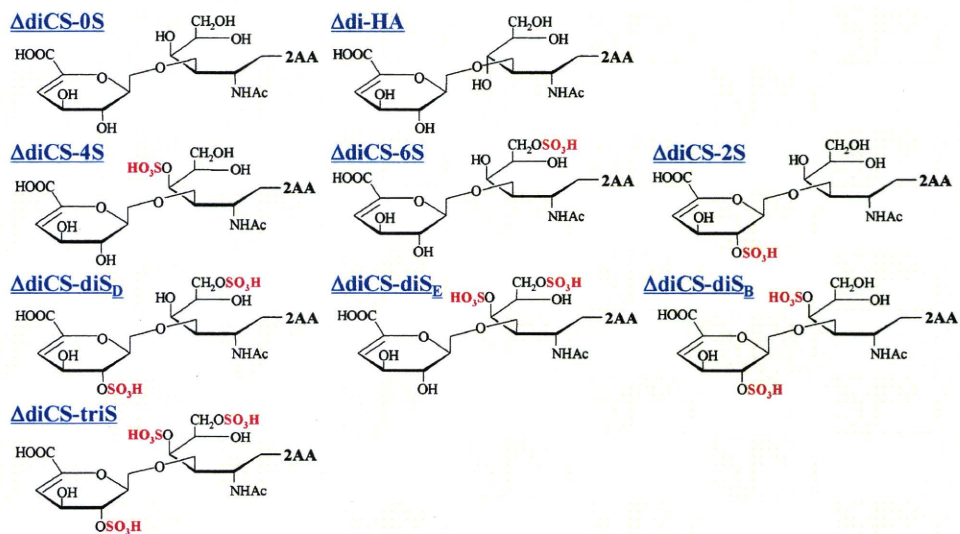


Fig.8 CE analysis of unsaturated disaccharides from HCT116 cell. Analytical conditions; Capillary, fused silica (40 cm x 50 μm.i.d). Buffer, 100 mM Tris-phosphate buffer (pH 3.0). Applied voltage, 25 kV. Injection, pressure method (1.0 psi, 10 sec). Temperature, 25 °C. Detection, He-Cd laser induced fluorescent detection (Ex: 325 nm, Em: 405 nm).

CS and HA unsaturated disaccharides



HS unsaturated disaccharides

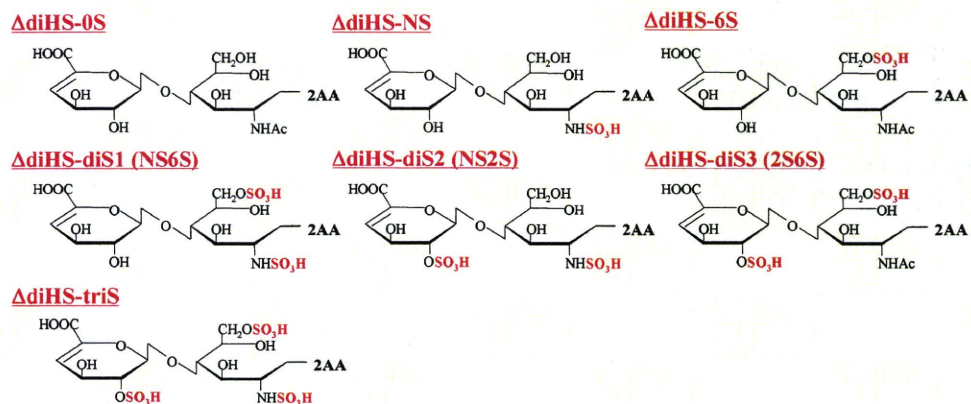


Fig.9 Structures of unsaturated disaccharides from CS and HS.

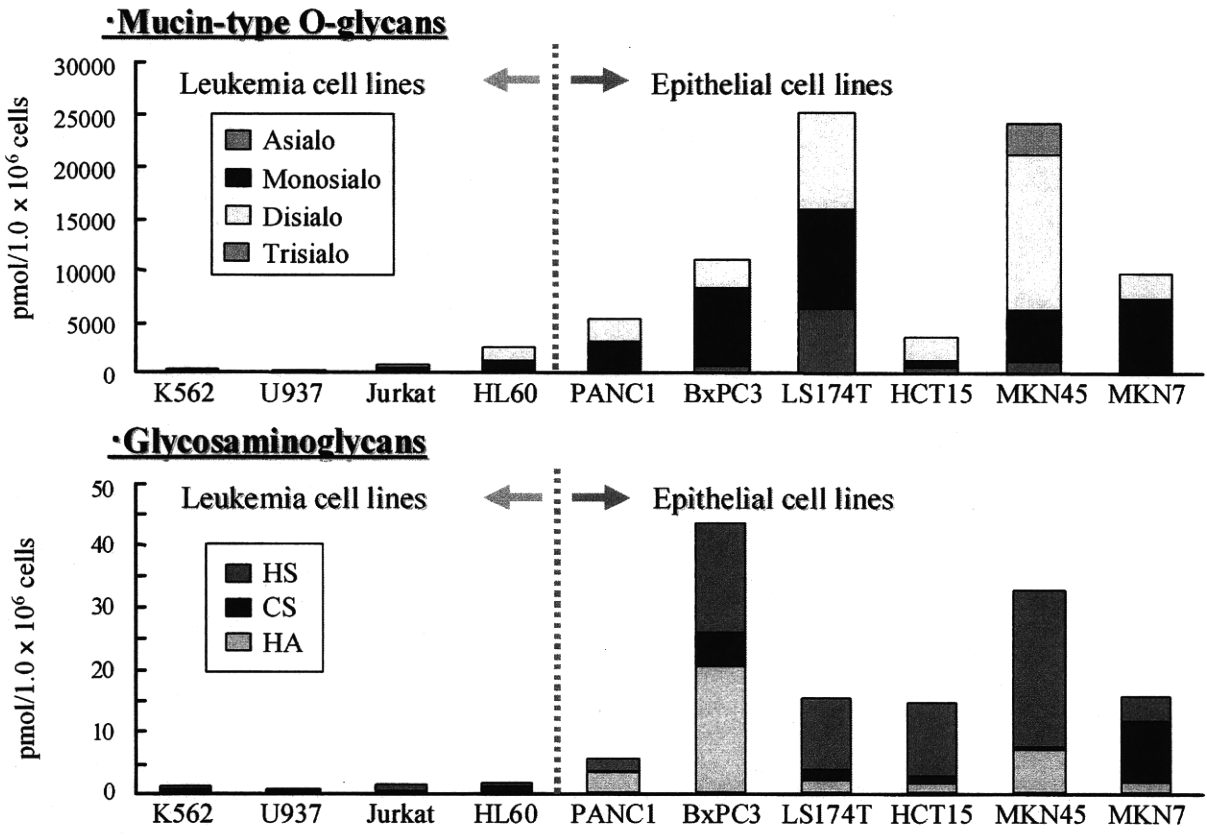
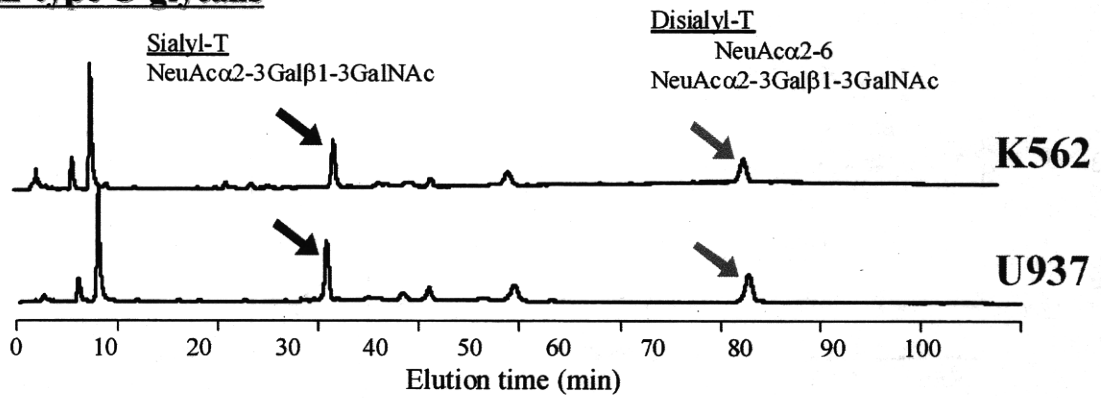


Fig.10 Comparison of the amounts of O-glycans expressed on cancer cells.

Mucin-type O-glycans



Glycosaminoglycans (CS)

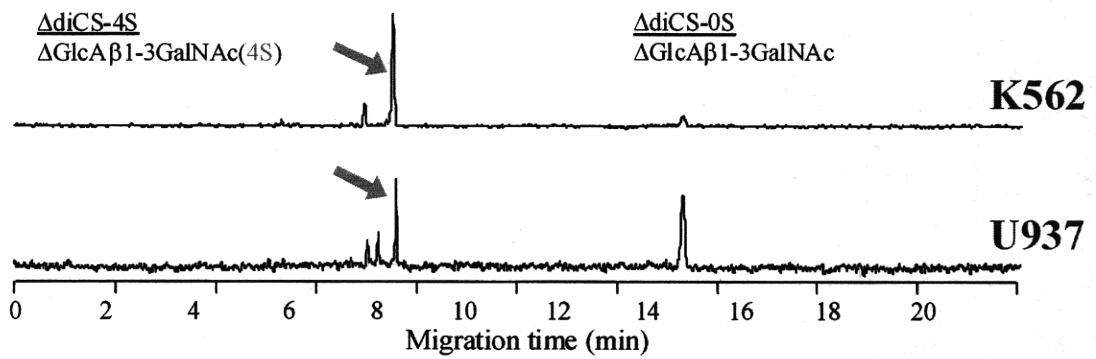
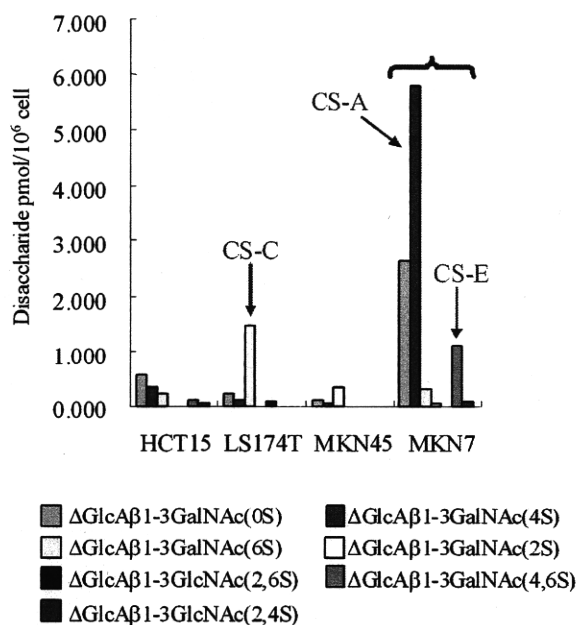


Fig.11 CE analysis of unsaturated disaccharides from K562 and U937 cell. Analytical conditions; Capillary, fused silica (40 cm x 50 μ m.i.d). Buffer, 100 mM Tris-phosphate buffer (pH 3.0). Applied voltage, 25 kV. Injection, pressure method (1.0 psi, 10 sec). Temperature, 25 $^{\circ}$ C. Detection, He-Cd laser induced fluorescent detection (Ex: 325 nm, Em: 405 nm).

CS/DS Disaccharides



HP/HS Disaccharides

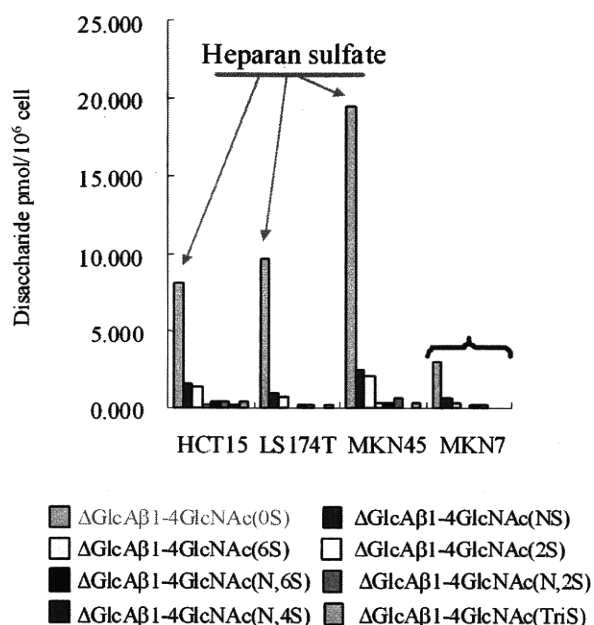
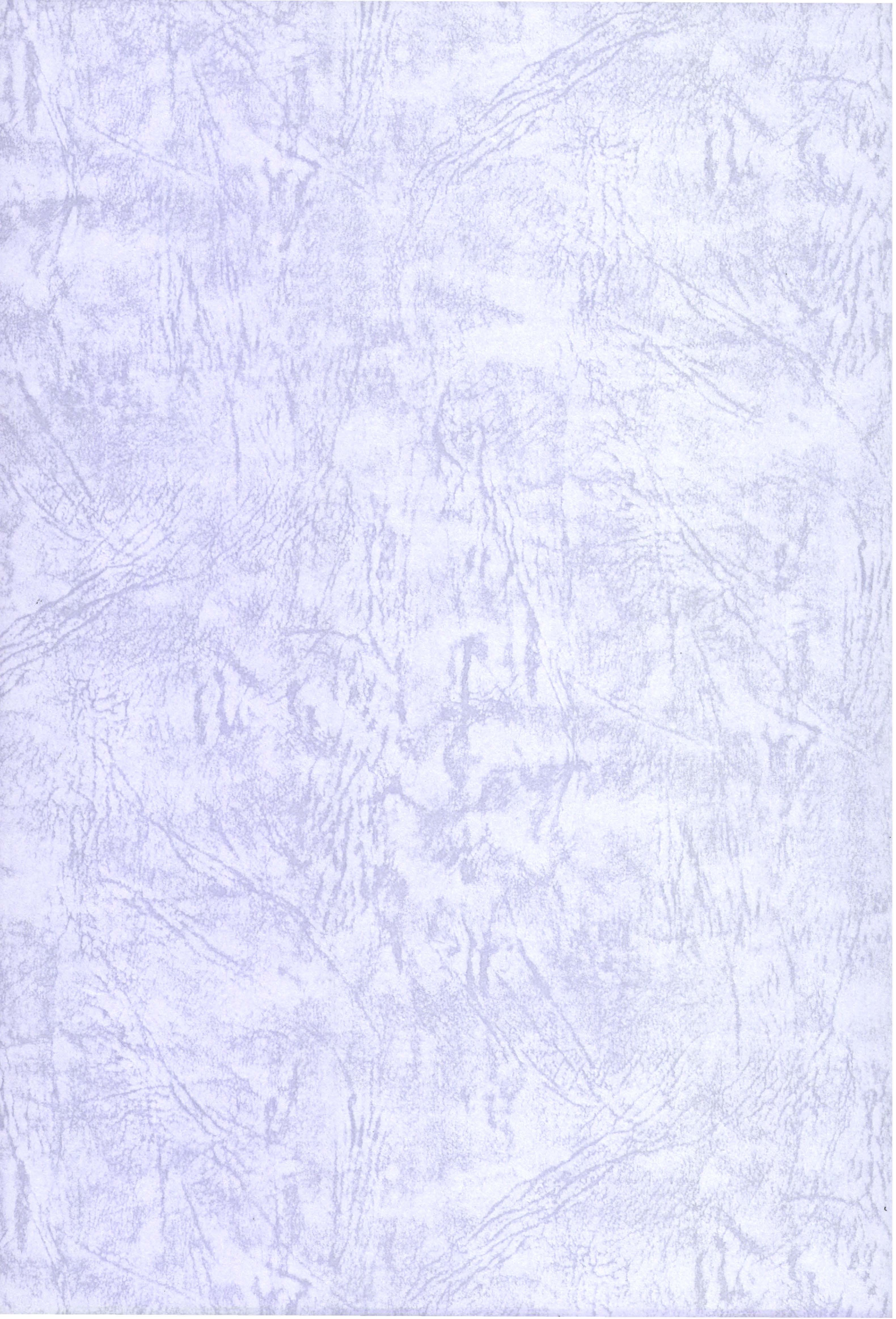


Fig.12 Comparison of the GAGs expressed on cancer cells



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