Electronic Supplementary Material (ESI) for Analyst.

Supplementary Table S1a. Extracted ions for disaccharide Gal-4-GlcNAc (± 0.15 Da).

Glycan Nodes	Extracted Ions (m/z)
t-Gal	59.05+71.05+87.04+101.06+117.05+129.05+145.08+161.07+173.08+205.10
4-Gal	59.05+71.08+87.04+101.06+113.06+117.05+129.05+131.07+161.07+173.07 +233.11
2-Gal	59.05+71.05+87.04+101.06+113.05+117.05+129.05+145.08+161.08+189.07 +205.10
3-Gal	59.05+74.04+87.06+101.06+117.05+129.05+143.08+161.08+173.07+201.08 +233.11
6-Gal	59.05+71.08+87.04+99.04+101.06+117.05+129.05+159.06+161.08+173.07+ 189.07+233.11
3,4-Gal	59.05+74.04+87.04+117.05+129.05+143.08+161.08
2,3-Gal	86.07+129.05+143.08+161.08+201.08+261.10
3,6-Gal	98.07+101.06+117.05+129.05+189.07
4-GlcNAc	74.06+98.06+116.04+142.08+158.08+233.11
3,4-GlcNAc	74.06+98.06+116.07+142.08+158.08
4,6-GlcNAc	74.06+98.07+116.07+142.08+158.08

Supplementary Table S1b. Extracted ions for disaccharide GalNAc-3-Gal (± 0.15 Da). Both furanose and pyranose form of 3-Gal (3-Gal *f* and 3-Gal *p*) were measured and summed to obtain a total signal for 3-Gal.

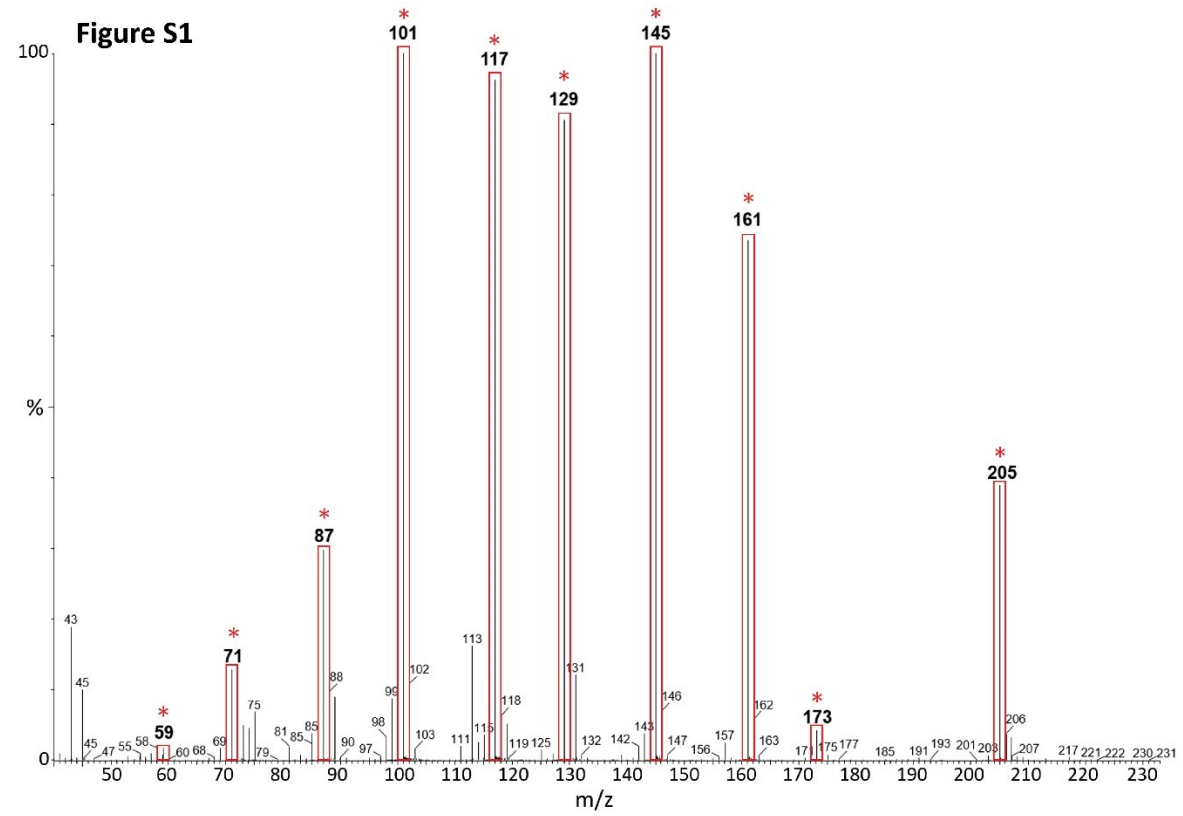
Glycan Nodes	Extracted Ions (m/z)
3-Gal f	59.05+74.04+87.05+101.06+117.06+161.09+171.07+189.09
3-Gal p	59.05+74.04+87.05+99.05+101.06+117.06+129.06+143.08+161.09+173.09+ 201.08+233.11
3,4-Gal	87.05+99.05+101.06+117.06+129.06+143.07+161.09
2,3-Gal	86.07+129.06+143.08+161.08+201.08+261.11
3,6-Gal	98.05+101.05+117.06+129.06+189.06
t-GalNAc	74.06+87.05+98.06+101.06+116.08+129.06+145.09+158.09+205.12
3-GalNAc	99.09+116.08+129.05+158.09+171.09+197.09
4-GalNAc	98.07+116.08+129.05+158.09+233.11
6-GalNAc	74.06+98.07+116.08+129.05+158.09+189.09+203.1
3,6-GalNAc	74.06+98.07+116.08+129.05+158.09+171.09+189.09

Supplementary Table S2a. Masses (as $[M+Na]^+_{avg}$) of intact O-glycans from fetuin.

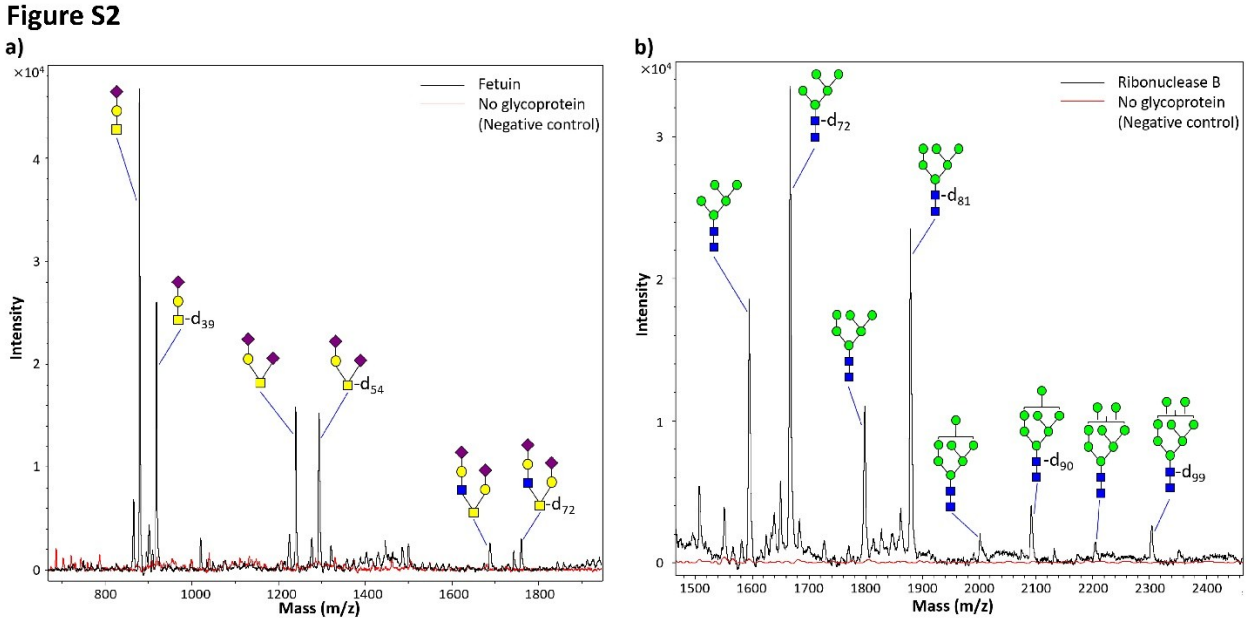
Labeling Reagent	Trisaccharide	Tetrasaccharide	Hexsaccharide
Iodomethane	$C_{38}H_{68}N_2O_{19}$	$C_{54}H_{95}N_3O_{27}$	$C_{74}H_{130}N_4O_{37}$
	$[M+Na]^+$: 879.94	$[M+Na]^+$: 1241.33	$[M+Na]^+$: 1690.82
Iodomethane-d3	$C_{38}H_{29}D_{39}N_2O_{19}$	$C_{54}H_{41}D_{54}N_3O_{27}$	$C_{74}H_{58}D_{72}N_4O_{37}$
	$[M+Na]^+$: 919.18	$[M+Na]^+$: 1295.66	$[M+Na]^+$: 1763.26

Supplementary Table S2b. Masses (as $[M+Na]^+_{avg}$) of intact N-glycans from ribonuclease B.

Labeling Reagent	$Man_5GlcNAc_2$	$Man_6GlcNAc_2$	$Man_7GlcNAc_2$	$Man_8GlcNAc_2$
Iodomethane	$C_{70}H_{128}N_2O_{36}$	$C_{79}H_{144}N_2O_{41}$	$C_{88}H_{160}N_2O_{46}$	$C_{97}H_{176}N_2O_{51}$
	$[M+Na]^+$: 1596.75	$[M+Na]^+$: 1800.97	$[M+Na]^+$: 2005.19	$[M+Na]^+$: 2209.41
Iodomethane-d3	$C_{70}H_{56}D_{72}N_2O_{36}$	$C_{79}H_{63}D_{81}N_2O_{41}$	$C_{88}H_{70}D_{90}N_2O_{46}$	$C_{97}H_{77}D_{99}N_2O_{51}$
	$[M+Na]^+$: 1669.19	$[M+Na]^+$: 1882.47	$[M+Na]^+$: 2095.75	$[M+Na]^+$: 2309.02

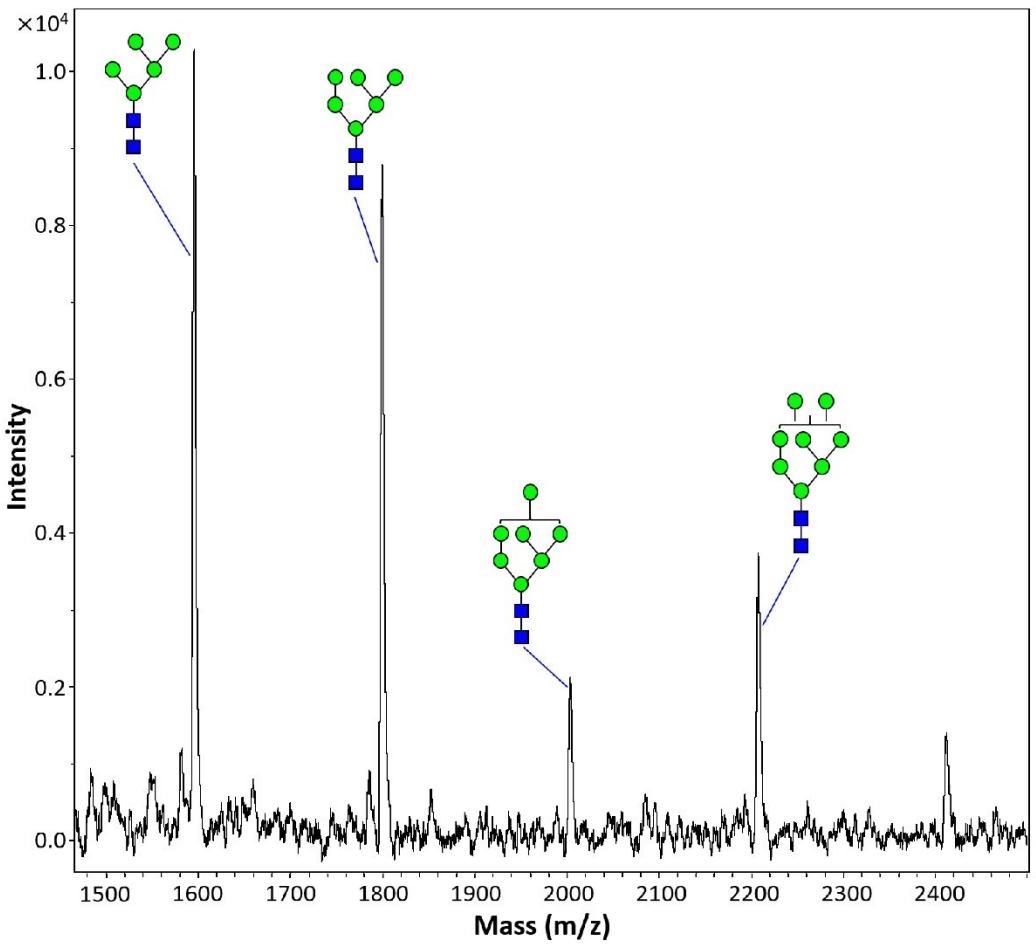


Supplementary Figure S1. Illustration of extracted ions for the PMAA representing terminal galactose (t-Gal). Spectrum was collected from a chromatographic peak with a retention time at 5.16 min.



Supplementary Figure S2. Overlaid spectra of intact glycans and negative controls. a) O-glycans of fetuin from fetal bovine serum overlaid with a negative control spectrum from a sample lacking glycoproteins. b) N-glycans of bovine Ribonuclease B overlaid with a negative control spectrum from a sample lacking glycoproteins. Glycan structure symbols indicate the

most likely (isomerically ambiguous) corresponding glycan permethylated by the SC 50 min procedure with unlabelled iodomethane, or by the SCF procedure with iodomethane-d_3 (as indicated by "-d_n" labels where the subscripts represent number of hydrogen atoms replaced by deuterium atoms in each glycan structure).



Supplementary Figure S3. MALDI-MS spectra of the four glycans released from 100 ng of bovine RNase B. The signal-to-noise (S/N) ratios of the peaks corresponding to all four glycans were greater than five.