

Integrated Microfluidic Lectin Barcode Platform for High-Performance Focused Glycomic Profiling

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Table S1. List of Lectins used in this work and their glycan binding specificities.

	Lectins	Origin	Binding specificity
1	PNA	<i>Arachis hypogaea</i>	Gal β 1-3 GalNAc α -Thr/Ser (T antigen)
2	LCA	<i>Lens culinaris</i>	Fuc α 1-6GlcNAc and α -Man, α -Glc
3	SBA	<i>Glycine max</i>	Terminal GalNAc (especially GalNAc α 1-3Gal)
4	AAL	<i>Aleuria aurantia</i>	Terminal α Fuc and \pm Sia-Le ^x
5	MAL II	<i>Maackia amurensis II</i>	Sia α 2-3Gal
6	SNA	<i>Sambucus nigra</i>	Sia α 2-6Gal/GalNAc
7	PHA-L	<i>Phaseolus vulgaris</i>	Tri- and tetra-antennary complex oligosaccharides
8	ECA	<i>Erythrina cristagalli</i>	Lac/LacNAc
9	RCA120	<i>Ricinus communis</i>	Galactose, Lac/LacNAc
10	DSL	<i>Datura stramonium</i>	(GlcNAc) _n , polyLacNAc and LacNAc (NA3, NA4)
11	GSL-II	<i>Griffonia simplicifolia</i>	Agalactosylated N-glycan
12	ConA	<i>Canavalia ensiformis</i>	α -Man (inhibited by presence of bisecting GlcNAc), α -Glc, complex-type N-glycans
13	GNA	<i>Galanthus nivalis</i>	non-substituted α 1-3 and α 1-6 Man
14	UEA-I	<i>Ulex europaeus</i>	Fuc α 1-2LacNAc
15	VVL	<i>Vicia villosa</i>	α -, β -linked terminal GalNAc and GalNAc α -Thr/Ser (Tn)
16	WGA	<i>Triticum unlgaris</i>	GlcNAc β 1-4GlcNAc β 1-4GlcNAc, β -GlcNAc and multivalent Sia

Table S2. Datasheet of three commercial CA125 protein samples used in this work

Vendor	MeridianLifeScience	MeridianLifeScience	Fitzgerald Industries
Catalog No.	A86928H	A97180H	30-AC21
Source	Human ovarian adenocarcinoma tissue	Ovarian carcinoma cell line	Ascitic fluids from ovarian cancer patients
Purity/ Purification	>50% pure by SDS-PAGE	Known Contaminants: CA19-9= 124 Units/mL(<0.1%) CA15-3= 2.15 Units/mL(<0.1%)	>95% pure
Concentration	650,000 Units/mL	83,840 Units/mL	750,000 Units/mL
Format & Buffer	Liquid in PBS (pH7.4) with 3% sucrose and 0.05% NaN ₃	Liquid in PBS (pH7.4 ± 0.2) with 0.1% NaN ₃	Liquid in PBS (pH7.2) with 3% sucrose and 0.05% NaN ₃

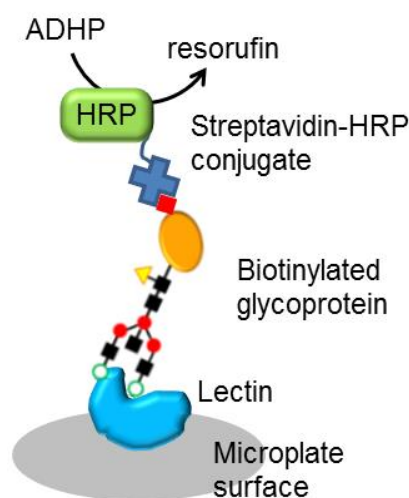


Figure S1. Scheme of the microplate-based lectin assay using horseradish peroxidase-conjugated streptavidin and the QuantaRed HRP substrate (ADHP) for absorbance or enhanced chemifluorescent detection.

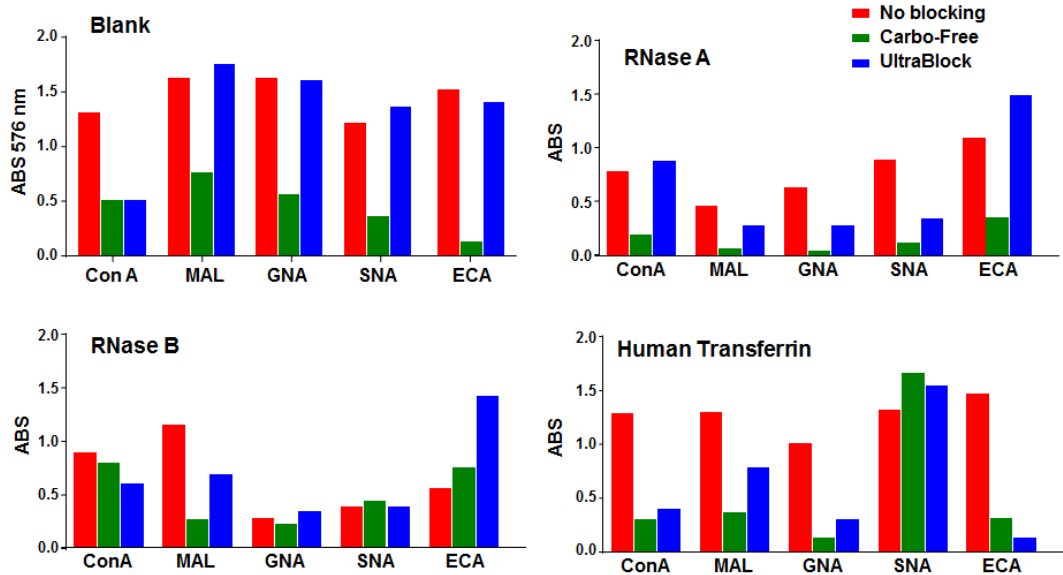


Figure S2. Effects of different blocking solutions, Carbo-Free (Vector) and UltraBlock (AbD Serotec), on the background and binding of two biotinylated glycoproteins and one control protein (1 μ M) using six lectins deposited in a 384-well microtiter plate.

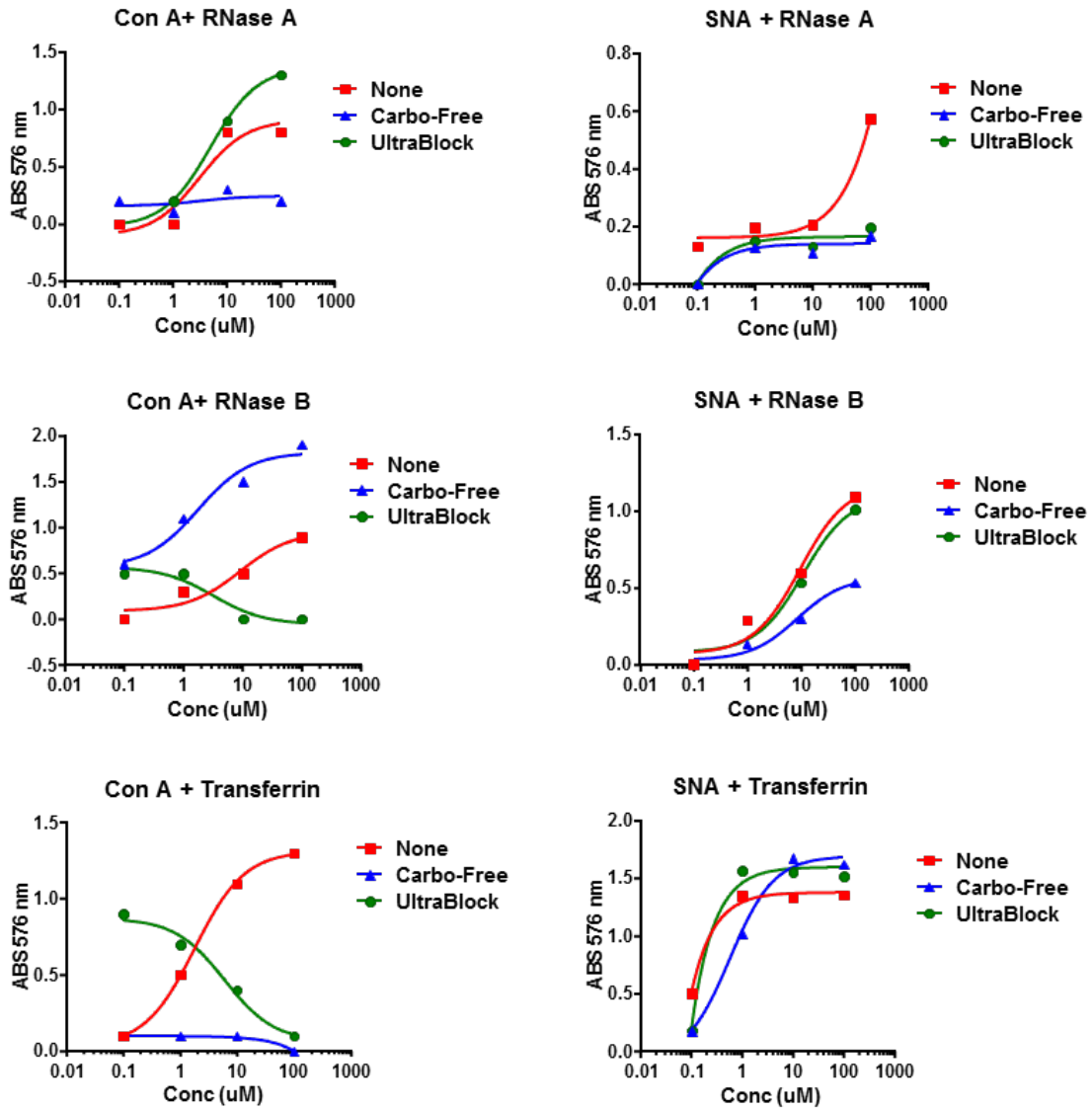


Figure S3. Concentration titration of the standard protein/glycoproteins using Con A and SNA confirms the blocking effectiveness of the Carbo-Free solution and suggests no inhibition on lectin-glycan binding, in contrast to the UltraBlock solution.

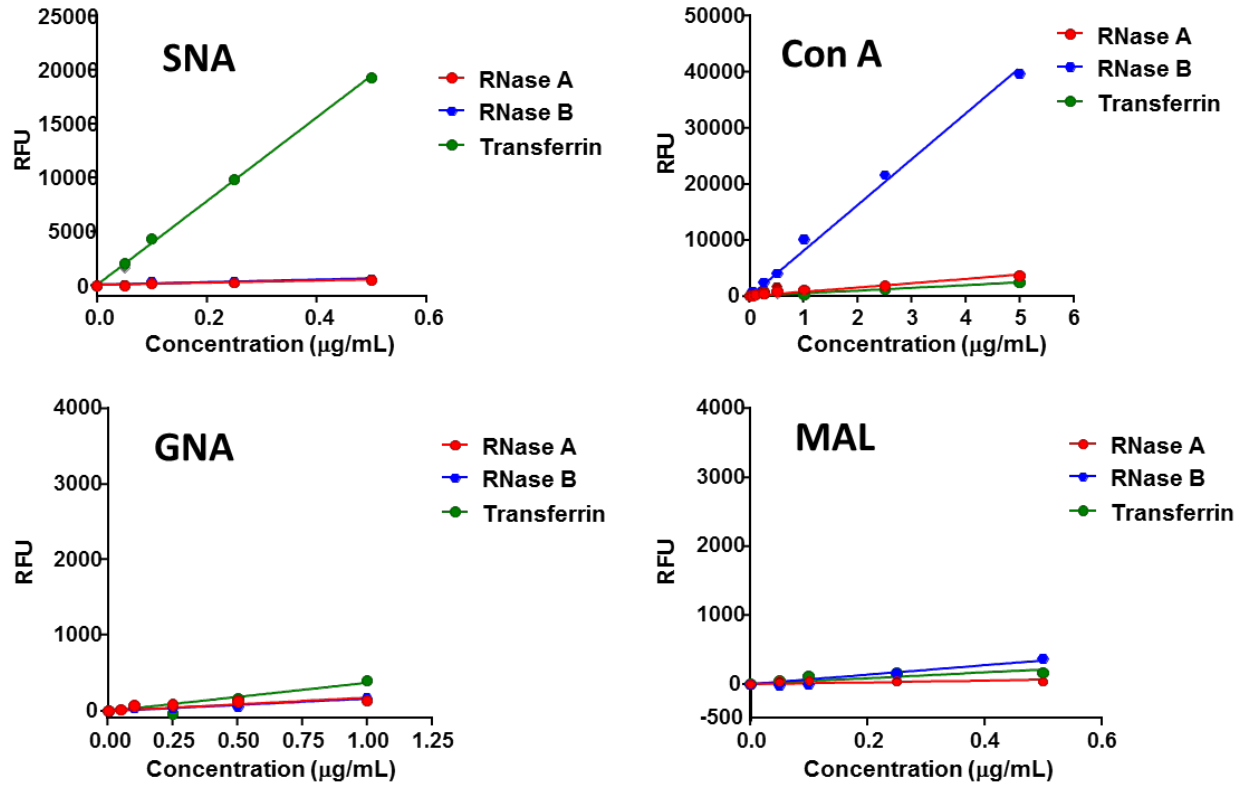


Figure S4. Quantitative detection of three standard protein and glycoproteins using various lectins. Assay conditions: buffer, TBST (20 mM Tris (pH 7.6), 150 mM NaCl, and 0.05% Tween 20), blocking buffer, Carbo-Free; HRP-STV, 1.5 mg/ml; overnight binding; QuantaRed fluorescence detection.

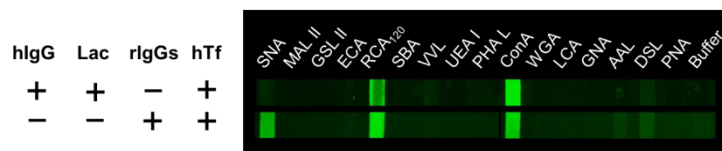


Figure S5. Comparison of blocking performance of a mixture of hIgG and lactose (top) and rIgG (bottom) for glycan profiling of hTf. The hIgG and lactose mixture blocker inhibits specific glycan binding for SNA and AAL.