Supporting Information

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Assessing the terminal glycosylation of a glycoprotein by naked eye

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Experimental Section
General procedures are as described[6]. “Buffer” in the following refers to 0.1 M citrate/phosphate buffer, pH 5.0.

Synthesis of TMR-B. 6-Tetramethylrhodamine isothiocyante (Molecular Probes, 7.4 mg, 17 µmol) was added to a solution of (5-amino-2-hydroxymethylphenyl)boronic acid (HCl salt) dehydrate (3.1 mg, 17 µmol, Combi-Blocks Inc.) in 0.1 M NaHCO₃/Na₂CO₃, pH 9 (1 mL) and DMF (1 mL). The mixture was stirred over night at rt in the dark. The mixture was concentrated under vacuum and the crude product was purified by adsorption on a Sep-Pak (C-18) cartridge, washed with H₂O, and eluted with 30-50% MeCN in H₂O (containing 10% 0.1 M HCl) giving a purple solid (6.3 mg, 64%). ES-MS m/z found 593.2 ([M]+, calcld 593.2).

Preparation of CPG-O-NH₂. Aminopropyl controlled pore glass (AMP-CPG, 1400Å pore-size, Millipore) (500 mg, loading of amino groups: ~ 50 µmol/g) was washed with DMF (3 x 2 mL), 50% diisopropylethylamine (DIPEA) in DMF (3 x 2 mL) and DMF (3 x 2 mL). The beads were treated with a mixture 4-N-Fmoc-aminooxymethyl-benzoic acid carboxymethyl ester [compound 14, reference 6b] (45 mg, 100 umol), O-(Benzotriazol-1-yl)-N,N,N',N'-tetramethyluronium tetrafluoroborate (TBTU, 25 mg, 75 µmol) and DIPEA (13 µL, 75 µmol) in DMF (2 mL) for 2 h at rt. The resin was washed with DMF (3 x 2 mL) and DCM (3 x 2 mL). Unreacted amino groups were capped by treating the beads with 50% Ac₂O in pyridine (2 mL) for 15 min at rt. Silanol groups (on the glass surface) were capped by treating the beads with 5% dichlorodimethylsilane in toluene (2 mL) for 30 min at rt, followed by 10 min incubation in dry MeOH. The silanol capping was repeated followed by washing with MeOH, DMF and DCM. A small portion of the resin was removed and the loading was determined to be approximately 35 µmol/g by comparison with a standard curve after release of the Fmoc-group.
The following manipulations were performed under a stream of argon to exclude laboratory air that may contain interfering small-molecule carbonyl compounds. Just prior to sugar capture, the capped Fmoc-protected beads (10 mg) were placed in a 1 mL plastic syringe equipped with a Teflon filter and mounted on a vacuum manifold. The Fmoc-deprotection was accomplished by treatment with 10% DBU in DMF (200 µL) for 20 min. The beads were then washed with DMF (3 x 200 µL) and freshly-prepared buffer (3 x 200 µL) to give CPG-O-NH$_2$, ready for capture experiments. Use of citrate-phosphate buffers that had been stored several weeks resulted in an increase in background fluorescence from an acceptable 50 RFU (in a typical experiment) to over 350 RFU.

**Preparation of β-galactosidase.** A solution of β-galactosidase (Sigma, G4142, 100 µL, 0.52 mg enzyme/mL, 0.10 U) was diluted with buffer (2 mL) and placed in a pre-washed Vivaspin 2 mL concentrator (10000 MWCO RC). The assembled concentrator was centrifuged (3 min at 4 °C and 4,000 x g), refilled with buffer and centrifuged again (4 x 2 mL for 3 min at 4 °C and 4,000 x g). The concentrator was inverted and centrifuged (2 min at 4 °C and 3,000 x g) giving enzyme in the filtrate cup (38 µL, ca. 2.6 U/mL).

**Preparation of asialo fetuin (A-Fet).** A solution of A-Fet (from fetal calf serum, Type I, Sigma, A4781, 500 µL of a 1 mg protein/mL buffer) was exchanged vs. buffer as described for the β-galactosidase above to provide 100 µL of solution, ca. 5 mg/mL in protein.