Polymer-Functionalised Surfaces for Microarray Applications

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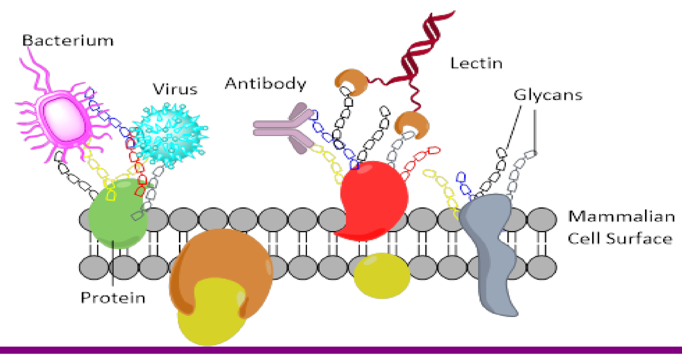
1. Carbohydrate Microarrays

- Global decrease in new antibiotics and an increase in antibiotic resistance
- Need for new technologies to investigate infection and rapidly diagnose
- Prior to infection, pathogens must typically adhere onto host cells through protein-carbohydrate interactions



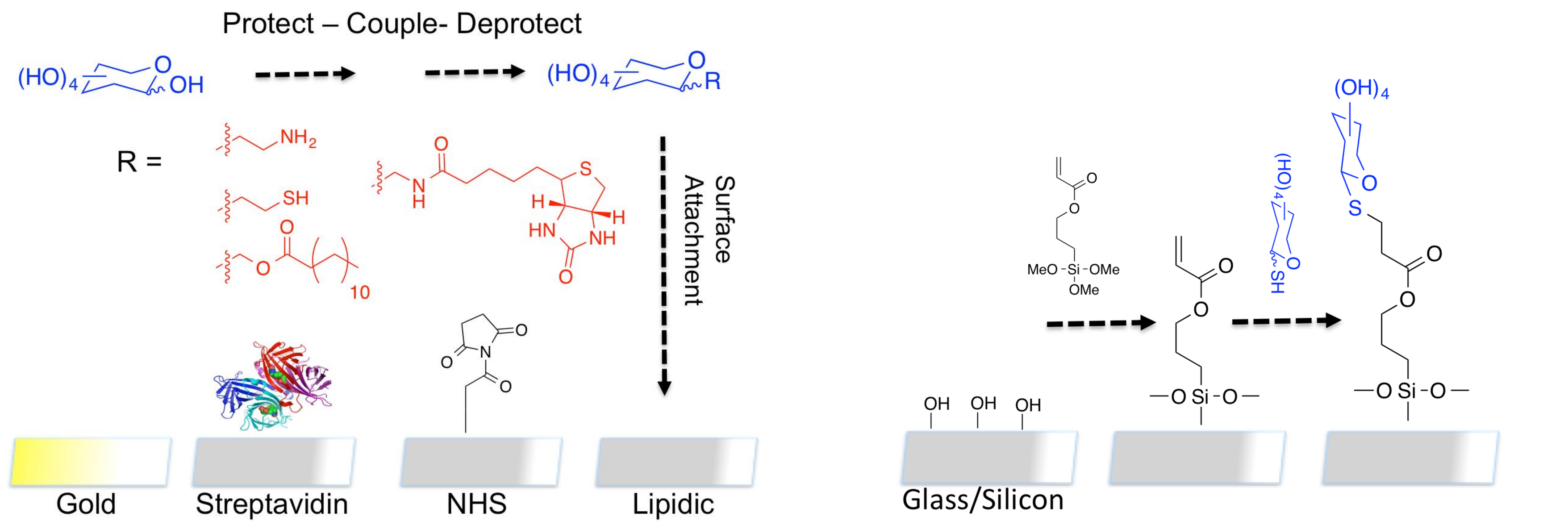
Microarrays can be used to:

- Probe protein-carbohydrate interactions in a high-throughput manner
- Provide structural information on pathogens and carbohydrates
- Aid development of anti-adhesion therapy
- Develop improved diagnostic devices

2. The need for Simple Carbohydrate Immobilisation Techniques

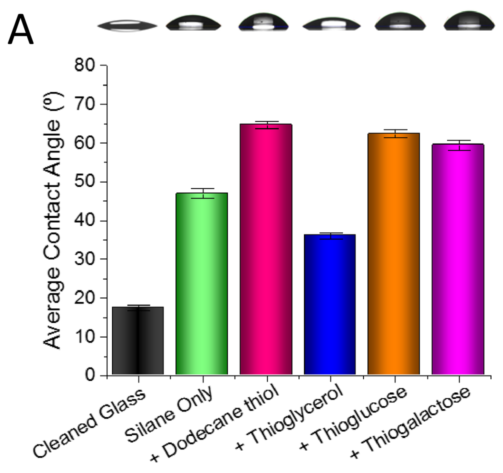
Immobilisation strategies to assemble carbohydrate arrays:

- **Non covalent** (e.g. lipid binding, strepdavidin) + Straightforward - Liable to washing-off effects
- **Covalent** (e.g. boronic acid, azidoaryl) + Stability - Multiple glycan orientations
- **Site specific covalent** (e.g. amino linkers with succinimidyl ester coatings) + Control and stability

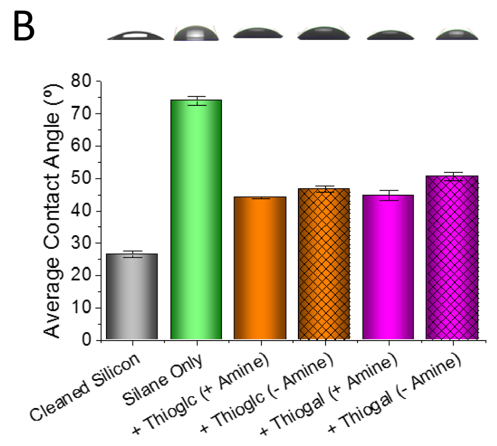


Our thiol-ene route using acrylate coated glass/silicon as a template surface for forming covalently tethered monolayers

A

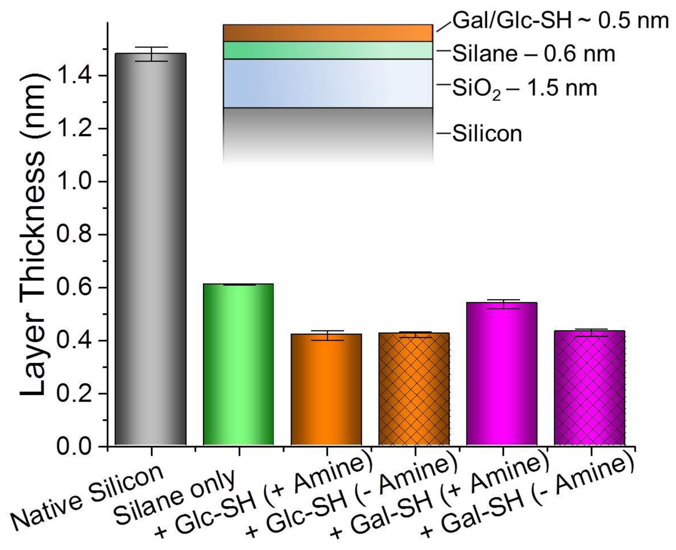


B



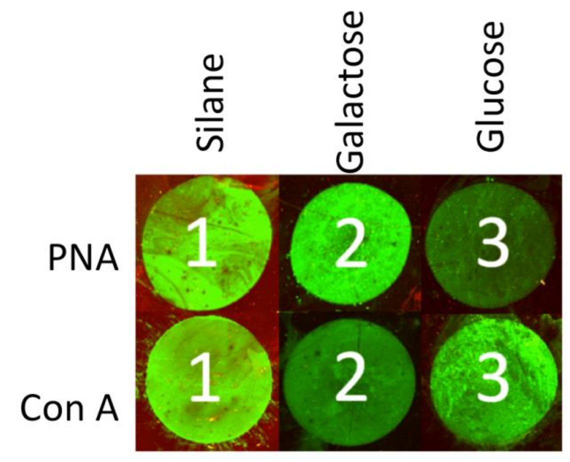
Contact angle measurements A) Glass slides; B) Silicon wafers

Ellipsometry measurements Demonstrating the thickness of each layer on the silicon wafers



Microarray applications

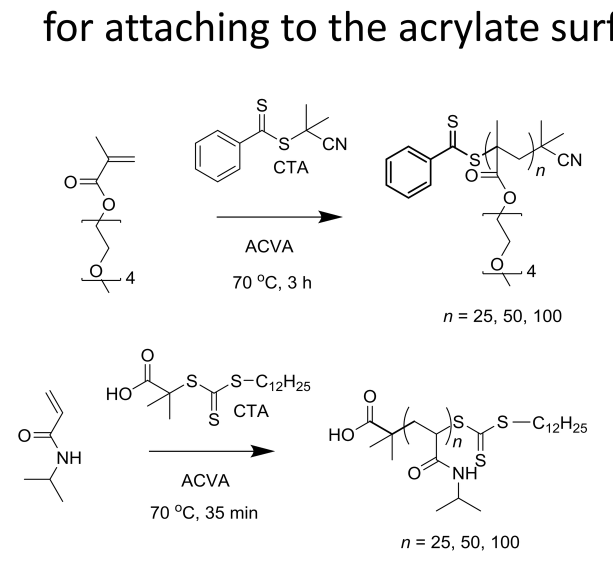
- Surfaces bearing either silane or Gal-SH/Glc-SH
- Exposed to fluorescent fluorescein-labelled lectins
- PNA (peanut agglutinin) and Con A (Concanavalin A)
- PNA prefers galactose
- Con A for glucose/mannose

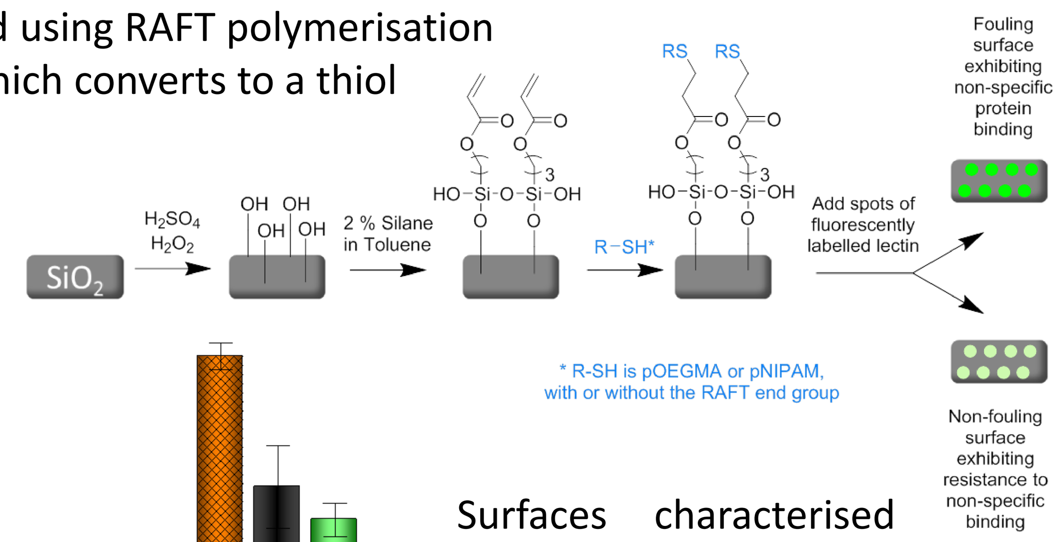


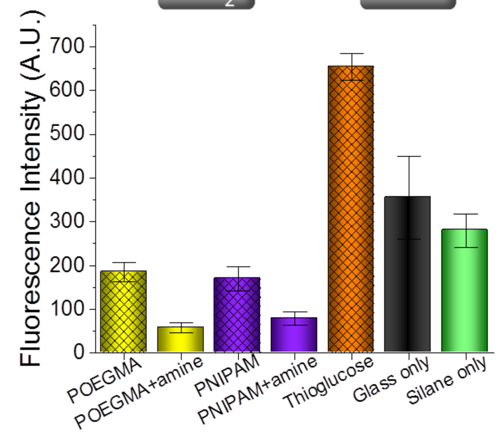
Biggs, C.I., Edmondson, S., Gibson, M.I., *Biomaterials Science* 2014, 3, 175–181

3. The Need for Arrays which Resist Non-Specific Adsorption

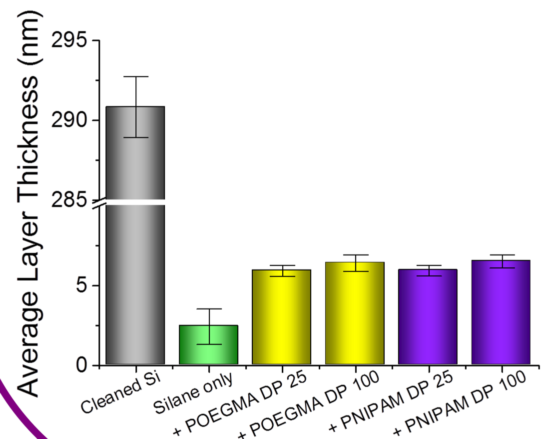
- Non-specific binding reduces resolution & gives false positives, polymer brushes can reduce this
- Polymers of **POEGMA** (poly[oligo(ethylene glycol methylether methacrylate)]) & **PNIPAM** (poly(*N*-isopropylacrylamide)) were synthesised using RAFT polymerisation
- Polymers contain a RAFT end group, which converts to a thiol for attaching to the acrylate surfaces

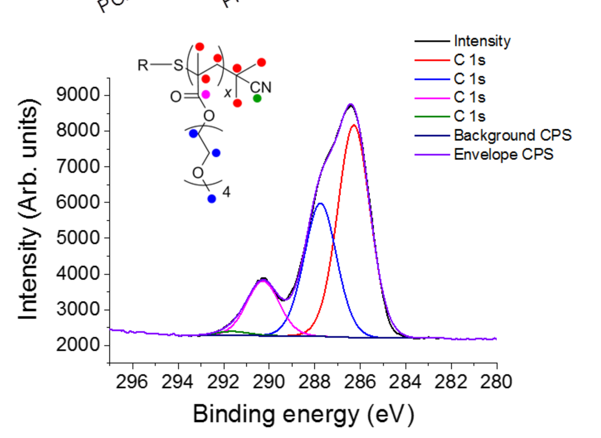


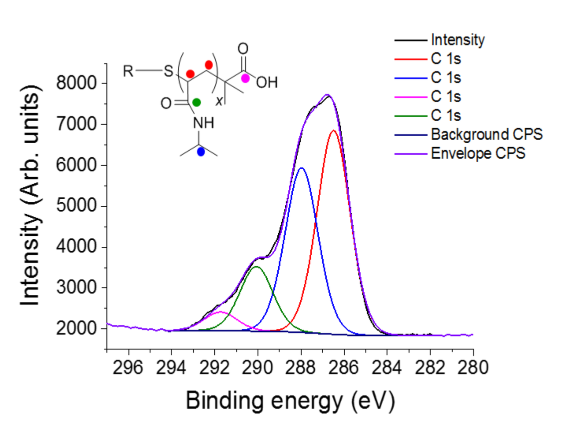




Surfaces characterised by ellipsometry, Contact Angle and XPS

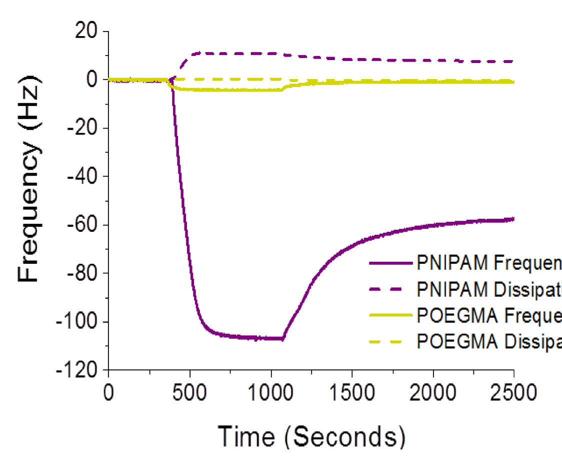


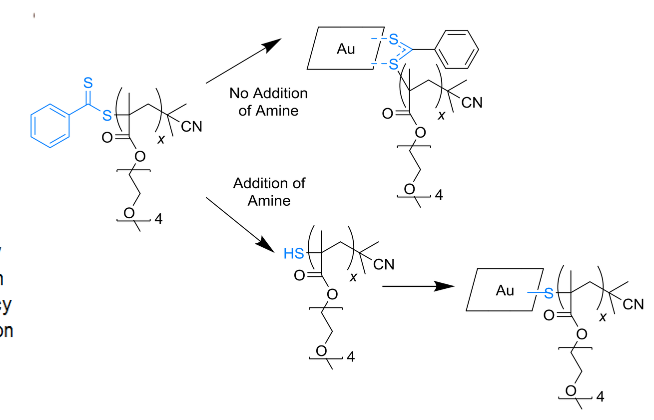


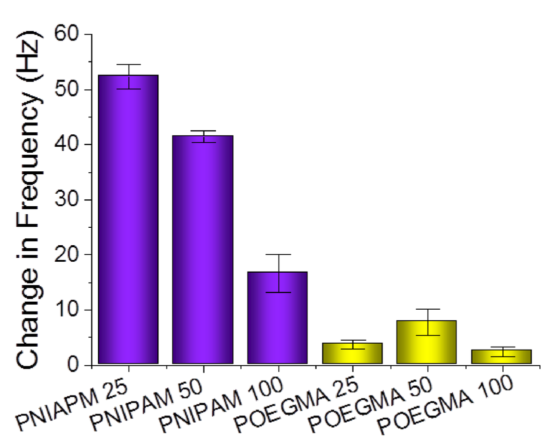


Quartz Crystal Microbalance Experiments

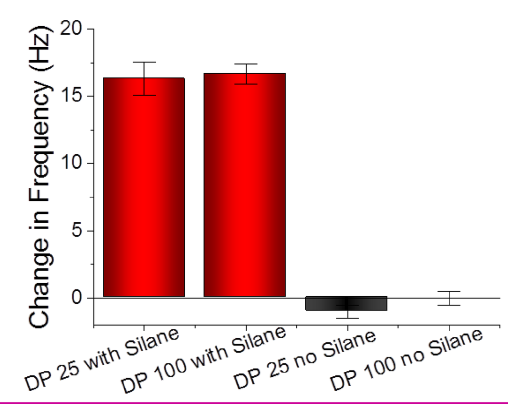
- Thiol-grafting to gold to compare grafting of PNIPAM and POEGMA was monitored in real time using a **Quartz Crystal Microbalance with Dissipation (QCM-D)**
- More PNIPAM bound to the surfaces than POEGMA, likely due to sterics

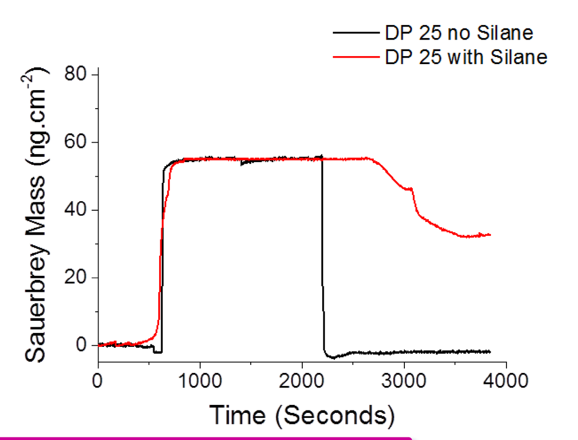


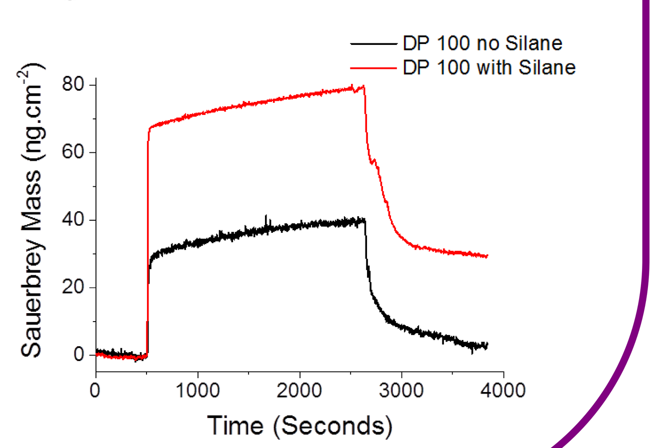




- Silicon QCM-D sensors as a direct comparison to our glass and silicon substrates
- Binding of PNIPAM only observed in presence of the silane coating



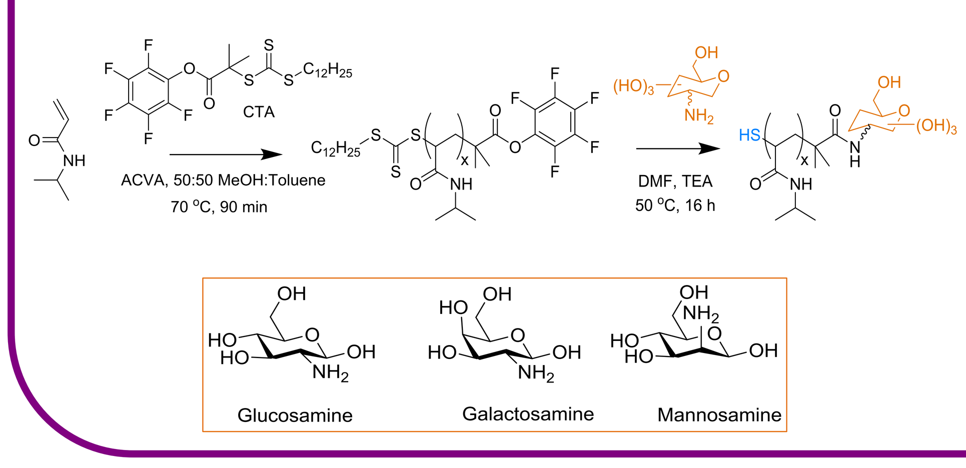




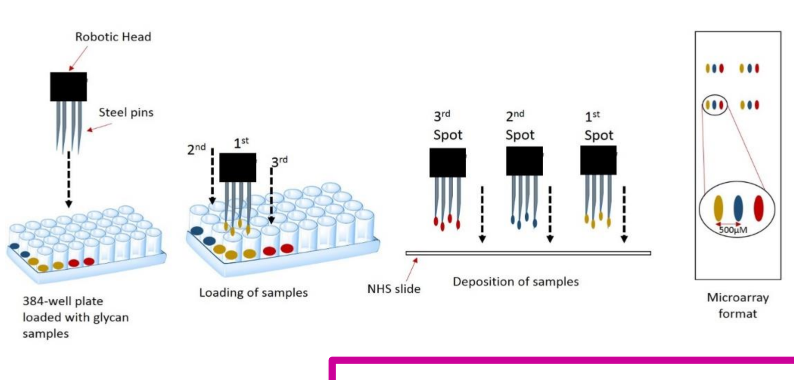
Biggs, C.I., Walker, M. Gibson, M.I., *Biomacromolecules* 2016, In Press

4. High-throughput Arrays of Surface Immobilised Glycopolymers

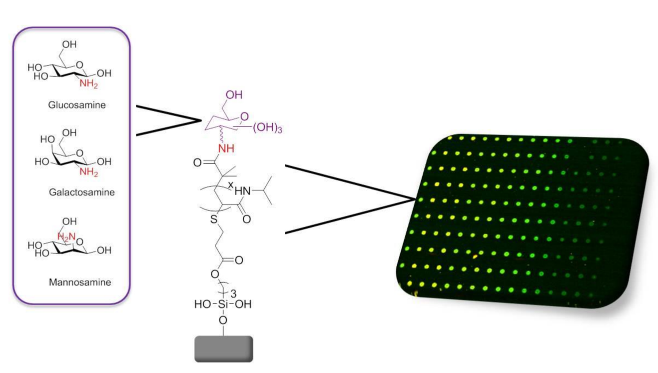
Synthesis of pNIPAM glycopolymers with RAFT end group (thiol) at one and glycan at the other



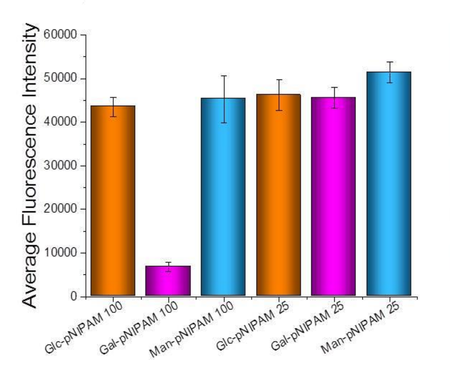
Direct microcontact printing of the glycan terminated polymers onto the acrylate functionalised glass slides

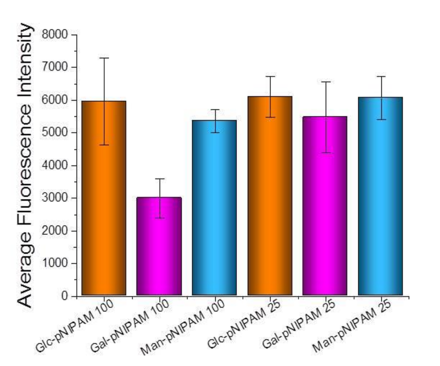


Generation of high-density arrays of surface immobilised glycans, with polymer tethers



Incubation with fluorescently-labelled lectins to assess protein-carbohydrate binding patterns





Biggs, C.I., Gibson, M.I., In Preparation

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