Understanding bio-nano interactions at the blood vessel wall

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Nanomaterial-cell interactions have been extensively explored as they are a precursor for nanomaterial uptake into cells. Crucially, all cell membranes are decorated in sugar macromolecules (glycans) which form a negatively charged matrix bound via proteins and lipids, referred to as the glycocalyx (**Fig. 1A**). Nanomaterials must pass through the glycocalyx prior to interactions at the cell membrane (**Fig. 1B**). However, we do not fully understand the impact of the glycocalyx on nanomaterial interactions at the cell surface. This is in large part due to most studies using isolated cells devoid of a glycocalyx[1].

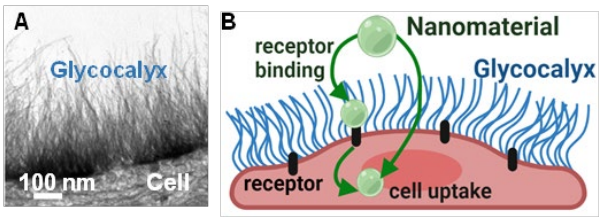

Figure 1: (A) The glycocalyx is a matrix of anionic sugar macromolecules anchored at the cell membrane[2]. (B) The glycocalyx influences cell-nanomaterial interactions critical for cell uptake.

Almost all nanomaterial drug delivery systems are transported from the place of administration to target sites via the circulation[2]. Thus, interactions with endothelial cells are essential for nanomaterial transport from the bloodstream to tissues prior to drug release. This study analysed interactions between the endothelial glycocalyx and poly(acetyl, arginyl) glucosamine (PAAG) polymers, which contain guanidino groups with high affinity for carboxylates and sulphates within glycans.

Primary human umbilical vein endothelial cells (HUVECs) were cultured over 7 days to produce a robust glycocalyx, including the glycans hyaluronan (HA) and heparan sulphate (HS), and exhibited mature cell-cell junctions expressing VE-cadherin. PAAGs were not cytotoxic to HUVECs at doses analysed up to 200 µg/mL and did not alter the barrier function properties of the cells in either transendothelial electrical resistance or transwell assays. PAAGs bound to the HUVEC glycocalyx and co-localised with HA and HS, and were internalised as established via lattice light sheet and fluorescence lifetime imaging microscopy. These data indicate the ability of cationic polymers to pass through the glycocalyx and be internalised.

The binding between PAAG and either HA or HS was measured by quartz crystal microbalance with immobilised thiolated glycans and PAAG exposed to this layer under flow to simulate interactions at the cell surface. This analysis revealed that the binding of PAAG to either HA or HS was initially flexible and became more rigid with increasing interaction time consistent with the formation of biomolecular condensates observed when PAAG and either HA or HS were mixed in solution and observed via phase contrast and confocal microscopy.

Together this study adds to our understanding of nanomaterial properties that facilitate binding to and transport through the glycocalyx for intracellular uptake. This knowledge is an important step toward the design of more efficient and effective nanomaterial drug delivery systems which can interact with target cells in a controlled and predictable manner.

References:

[1] Fu, L.; et al. *Adv Drug Deliv Rev* **2022**, *184*, 114195.
[2] Weinbaum, S.; et al. *Annual Rev Biomed Eng* **2007**, *9*, 121.