

CONTROL OVER PROTEIN ADSORPTION WITH THERMOSENSITIVE STIMULI-RESPONSIVE COATINGS

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INTRODUCTION: Control over the interactions between biological components and synthetic interfaces is one of the most important goals in biomedical research. This goal is shared by a range of disciplines including biomaterials and tissue engineering as well as biochips and diagnostic devices. Modification of surfaces with thin polymer coatings is commonly employed to control interfacial biological interactions for applications in biosensors, microarrays, cell sheet engineering and 'lab on a chip' devices.

Advances in these applications are expected from the development and integration of stimuli-responsive or switchable coatings that can provide attractive functional properties to manipulate specific biological responses such as adsorption/desorption of biomolecules. Switchable coatings show considerable promise for the realisation of spatial and temporal control over interactions with biomolecules such as proteins and DNA, as well as with cells and bacteria.

METHODS: As part of ongoing research in our lab we report here our findings on stimuli-mediated protein adsorption to thermosensitive poly(N-isopropylacrylamide) (pNIPAM) coatings in hydrated and collapsed states.

Coatings of pNIPAM were prepared on silicon wafer substrates which had first been functionalised with an alkoxy-silane bearing methacrylate groups. Silicon wafers thus functionalised with surface polymerisable groups were then grafted with pNIPAM via radical polymerisation. Surface analysis was carried out using X-ray photoelectron spectroscopy (XPS), atomic force microscopy (AFM) and time of flight secondary ion mass spectrometry (ToF SIMS).

Adsorption of the proteins lysozyme (Lys) and bovine serum albumin (BSA) was investigated using TOF SIMS, AFM and XPS.

RESULTS: Surface characterisation of pNIPAM coatings by XPS showed elemental ratios matching values expected from the theoretical bulk polymer composition. The absence of Si in XPS survey

spectra indicated that the dry coatings were thicker than the XPS sampling depth of ~10 nm. In colloid probe (CP) AFM experiments, steric repulsion was observed at 4 times greater distance at $T < LCST$ than for $T > LCST$, indicating the swelling/deswelling nature of the coating. Furthermore, CP-AFM experiments with protein-functionalised tips allowed the forces between pNIPAM coatings and proteins to be recorded over a range of temperatures.

Analysis by ToF SIMS following protein adsorption experiments demonstrated that pNIPAM coatings showed no measurable fouling at 20 °C whilst they were protein retentive at 37 °C.

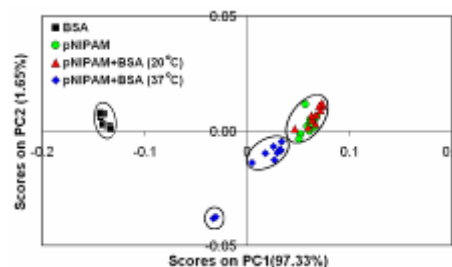


Fig. 1: Principal component analysis of positive ion ToF SIMS data. Score plot of PC1 vs PC2 for BSA, pNIPAM, pNIPAM + BSA (20 °C) and pNIPAM + BSA (37 °C) samples.

DISCUSSION & CONCLUSIONS: Surface coatings of pNIPAM were demonstrated to retain the polymer's inherent temperature-induced phase transition which was used to alter surface properties over modest temperature changes. Protein adsorption studies showed that pNIPAM coatings can be switched between non-fouling and fouling states in order to control protein immobilisation via small temperature changes. Protein could be adsorbed and desorbed reversibly.

The present study is expected to assist the development of switchable coatings for biomedical and biotechnological applications.

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