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Optimizing Protein Corona Recovery from AuNPs for MS Quantification

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Understanding protein–nanoparticle interactions is crucial for evaluating the behaviour of nanoparticles (NPs) in biological environments. While the physicochemical of NPs is determined by factors such as size, shape and surface chemistry, their biological identity is rapidly established upon exposure to biological fluids through the immediate adsorption of biomolecules such as proteins. In this work, our aim is to establish the foundation for a robust protein mass spectrometry (MS)-based quantification method to investigate the protein coronas formed on gold nanoparticles (AuNPs) of different sizes and surface coatings.
In addition, we aim to verify the successful formation of a protein corona after characterization by employing complementary analytical techniques, such as fluorescence-based tracking of labelled proteins, dynamic light scattering, and other solution-based assays. These steps allow us to validate that the adsorption of proteins has occurred prior to corona isolation.
Our approach relies on comprehensive particle system characterization, ensuring that observed changes in corona composition can be directly linked to well-defined nanoparticle properties. To monitor protein binding and recovery prior to MS analysis, we employed labelled proteins and defined test solutions as model systems for method development. A central challenge lies in designing an efficient protocol for protein corona isolation and desorption that minimizes loss while enabling recovery of the full complement of adsorbed proteins, with complete removal of NPs prior to MS. Looking ahead, the method is being tailored for compatibility with both bottom-up (peptide-based) and top-down (intact protein) MS approaches, providing a versatile framework for the quantitative and qualitative characterization of nanoparticle–protein interactions.

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