

## **Protein assembly onto polyelectrolyte multilayer films: a means to incorporate biological molecules into materials**

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The placement of biological or biomimetic structure onto or within synthetic materials portends significant advances in biosensing surfaces, drug delivery vehicles, and tissue engineering substrates. Multilayered nanofilms – formed by the electrostatically driven layer-by-layer surface adsorption of polyelectrolytes – are new and promising coatings onto (and into) which may be embedded functional biological molecules. Immobilization in multilayered films is simpler, more controllable, and more broadly applicable than conventional approaches such as chemical or affinity attachment or entrapment during material synthesis.

Fibronectin (Fn) is a matrix protein known to influence cell adhesion and spreading, wound healing, phagocytosis, and differentiation through its cell binding site (containing the RGD amino acid sequence). Materials coated with Fn are natural choices for tissue engineering substrates, but the optimal manner in which the protein is to be presented to a cell remains an open question. Ideal might be protein layers oriented in a way that maximizes exposure of the cell binding site and minimizes alteration of its internal conformation. Multilayered films, whose surface charge and structure may be controlled through processing variables, offer the possibility to bind Fn at an optimal orientation while preserving (and perhaps stabilizing) its internal conformation.

In this talk, we describe our recent work incorporating Fn onto and within polyelectrolyte multilayer films. Using optical waveguide lightmode spectroscopy (OWLS), we show multilayer film structure and formation kinetics to be greatly influenced by an applied electric field and subsequent Fn placement to be exquisitely sensitive to film structure and terminal layer charge. Using atomic force microscopy (AFM), we identify and quantify the tendency of Fn to aggregate on the multilayer film. By considering the electrostatic binding of a segmental model Fn, we explain the spreading of human umbilical vein endothelial cells seeded onto Fn/multilayer assemblies in terms of protein orientation and multilayer film structure.