Layer-by-layer assembly and disassembly of polymer and DNA films for gene delivery
Guangzhao Mao, Wayne State University, Detroit, MI 48202

Abstract

Layer-by-layer (LbL) films containing cationic polyelectrolytes and anionic bioactive molecules such as DNA are promising biomaterials for controlled and localized gene delivery for a number of biomedical applications including cancer DNA vaccine delivery. Bioreducible LbL films made of disulfide-containing poly(amido amine)s (PAAs) and plasmid DNA can be degraded by redox-active membrane proteins through the thiol-disulfide exchange reaction to release DNA exclusively into the extracellular microenvironment adjacent to the film. In order to better understand the film degradation mechanism and nature of the released species, the bioreducible film degradation is studied by atomic force microscopy, fluorescence, and dynamic light scattering in solutions containing a reducing agent. The PAA/DNA LbL film undergoes fast bulk degradation with micrometer-sized pieces breaking off from the substrate. This bulk degradation behavior is arrested by periodic insertions of a non-bioreducible poly(ethyleneimine) (PEI) layer. The LbL films containing PAA/DNA and PEI/DNA bi-layers display sequential film disassembly and are capable of continuously releasing DNA nanoparticles over a prolonged time. Insertion of the PEI layer enables the bioreducible LbL films to transfect human embryonic kidney 293 cells. The data conclude that the PEI layer is effective as a barrier layer against interlayer diffusion during LbL film assembly and more importantly during film disassembly. Without the barrier layer, the high mobility of cleaved PAA fragments is responsible for bulk degradation of bioreducible LbL films, which may prevent their ultimate gene-delivery applications. This work establishes a direct link among film internal structure, disassembly mechanism, and transfection efficiency. It provides a simple method to design bioreducible LbL films for sequential and long-time DNA release.