450g Quantum Dot-Peptide Assemblies for Anti-Prostate Cancer Therapy

Kaushal Rege and Martin L. Yarmush

Prostate cancer is the most common malignancy affecting men and is the second-leading cause of cancer death in men in the United States after lung cancer. The Prostate-Specific Membrane Antigen (PSMA) is a type II membrane glycoprotein which is over-expressed in all stages of human prostate cancer disease. Importantly, the expression of the protein increases in cases of hormone-refractory disease and metastasis. In these studies, we are focusing on the development of quantum dot-peptide assemblies as potential anti-prostate cancer therapeutics. Coiled-coil motifs are ubiquitous in biology and form the basis of molecular recognition and assembly in a variety of systems including fibrous proteins (e.g. keratin), transcription factors (e.g. yeast GCN4 leucine zipper), and viral proteins (e.g. gp41 of HIV). We first describe the design, recombinant expression, purification, and characterization of PSMA-targeting peptides (PTPs) using a coiled-coil based epitope display strategy. Different PTP mutants were generated using deletion/insertion site-directed mutagenesis and extensive biophysical characterization was carried out to obtain insights into the secondary structure and peptide stability in each case. We then investigated the in vitro efficacy of an apoptotic peptide using both, hormone-dependent (LNCaP) and hormone-refractory (PC3) prostate cancer cell lines. The mechanisms of peptide uptake by cells and the resulting cell death were investigated. In addition, liposome disruption / leakage experiments were carried out in order to investigate the membrane permeabilzation activity of the apoptotic peptide. The simultaneous display of a number of bioactive peptides on a scaffold is known to result in higher efficacies of the displayed peptide. We explored the use of quantum dots as scaffolds for the polyvalent display of the above peptides. In addition to the polyvalent display, the ability to image the fluorescent quantum dots is a dual advantage in potential delivery applications. Finally, we discuss the ‘bottom-up' assembly of these individual components leading to multi-functional constructs that possess targeting, apoptotic and imaging capabilities on a single platform.