Poly(methacrylic acid-g-ethylene glycol) Microparticles as Modulators of Tight Junction Morphology

Omar Fisher¹ and Nicholas A. Peppas¹,²,³

¹Department of Biomedical Engineering, University of Texas at Austin, Austin, Texas; ²Department of Chemical Engineering, University of Texas at Austin, Austin, Texas; ³Division of Pharmaceutics, University of Texas at Austin, Austin, Texas.

The hydrogen bonding disruption and pH-dependent swelling properties of poly(methacrylic acid-g-ethylene glycol) (P(MAA-g-EG) microparticles protect biomacromolecules from gastric degradation in oral protein delivery and facilitate their transport across the intestinal epithelium. Using measurements of transepithelial electrical resistance (TEER) and visualization of tight junction in vitro, here we examine the mechanism by which microparticles modulate transepithelial paracellular drug transport. The effect of (P(MAA-g-EG) with a 1:1 monomer ratio on the barrier function of the tight junction complexes and on the integrity of cell-to-cell adhesion was assessed using transepithelial resistance measurements coupled with immunofluorescent labeling. Caco-2 cell monolayers were cultured on 4 µm polycarbonate Transwell membranes for 21 days. The cells were then exposed to either phosphate buffered saline (PBS) with calcium (negative control), 10 mg/ml P(MAA-g-EG) microparticles in PBS without calcium, or ethylenediaminetetraacetic acid (EDTA) (positive control) in PBS for 4 hours with resistance measurements every 30 minutes. The membranes were then fixed and double stained using mouse anti-E-cadherin and rabbit anti-claudin primary antibodies followed by TRITC conjugated goat anti-mouse and FITC conjugated goat anti-mouse secondary antibodies. Laser scanning confocal microscopy was used to analyze the distribution of stained proteins within tight junctions following a 30% resistance drop across membranes exposed to microparticles. The results showed a morphological correlation between the stability of the tight junction complex and the decreased resistance following exposure to microparticles. The ability to observe cytoarchitectural changes associated with (P(MAA-g-EG) formulations allows for a more accurate monitoring of their therapeutic value as drug delivery vehicles.

Supported by grants from NIH and the NSF/IGERT Program