Microneedle-Mediated Transcutaneous Immunization with Plasmid DNA Coated on Cationic PLGA Nanoparticles Induces Strong Systemic and Mucosal Immune Responses

A. Kumar, Z. Cui
University of Texas at Austin

Purpose
The purpose of this study is to develop plasmid DNA-coated positively charged PLGA nanoparticles and to evaluate the ability of these nanoparticles to induce immune responses, including mucosal immunity, after they are administered onto a mouse skin area pretreated with microneedles.

Methods
Cationic PLGA nanoparticles were prepared by the nanoprecipitation-solvent evaporation method using PLGA and DOTAP. Plasmid DNA (pCMV-β or pCI-neo-sOVA) was coated on the surface of the cationic nanoparticles by gently mixing equal volumes of cationic nanoparticles with plasmid DNA. The mixing ratio was adjusted so that the final charge on the surface of the resultant nanoparticles is positive. Mice were immunized with the net positively charged DNA-coated nanoparticles transcutaneously on a skin area pretreated with microneedles. Antibody responses in serum, fecal, and bronchoalveolar lavage (BAL) fluid samples were determined using ELISA. Cellular immune responses were measured by ELISPOT using splenocytes isolated from the immunized mice.

Results
Mixing cationic PLGA nanoparticles with plasmid DNA at a 2:1 weight ratio produced net positively charged plasmid DNA-coated nanoparticles (particle size, 115 nm, zeta potential, 54 mV). Microneedle-mediated transcutaneous immunization with the net positively charged pCMV-β-coated nanoparticles induced a stronger immune response than transcutaneous immunization or intramuscular immunization with plasmid DNA alone. At a low dose (4 microgram DNA/mouse), the immune response induced by microneedle-mediated transcutaneous immunization with net positively charged pCI-neo-sOVA-coated nanoparticles was stronger than when the same DNA-coated nanoparticles were injected intramuscularly. Moreover, specific IgA responses were detected in the fecal samples and BAL fluid of the transcutaneously immunized mice, but not in the intramuscularly immunized mice. Finally, splenocytes isolated from the transcutaneously immunized mice also secreted higher levels of IFN-gamma, as detected using ELISPOT.

Conclusion
Transcutaneous immunization with net positively charged plasmid DNA-coated nanoparticles onto a skin area pretreated with microneedles induced strong systemic and mucosal immune responses.