THE HARTWELL FOUNDATION

2010 Individual Biomedical Research Award

Review of Proposed Research

Investigator: **Marie Egan, M.D.**
Associate Professor
Department of Pediatrics

Institution: **Yale University**

Proposal: **Synthetic Nanoparticles for Gene Correction of Cystic Fibrosis**

Dr. Egan proposes an innovative technology for the replacement of short fragments of DNA in the gene responsible for cystic fibrosis (CF), to correct a known mutation in this most common lethal inherited disorder in the Caucasian population. The disease is caused by mutations in the gene that encodes for the cystic fibrosis transmembrane conductance regulator (CFTR) protein, causing thick, sticky mucus to build up in the lungs, digestive system, and other organs of the body. Although average life expectancy is 37 years, 50% of affected individuals die in childhood from respiratory failure. Since identification of the CFTR protein more than 20 years ago, there has been intense interest in developing gene-replacement therapies to cure this deadly disease. In spite of significant scientific advances, all approaches have failed. Marie proposes an alternative strategy to repair the CF defect using peptide nucleic acids (PNA) designed to target specific sequences of the double stranded DNA in the CF gene and acting like *molecular scissors*, effectively cut out the defective segment of DNA and replace it with the proper sequence. To enable the PNA to effectively reach their target in the cell's nucleus, Marie plans to encapsulate them in specialized nanoparticles made from FDA approved biocompatible and biodegradable polymers. Her approach will be to target position 508 of the gene for CFTR, in the region where 90% of all CF mutations are known occur. In effect, PNA will interact with the CF cellular DNA and short strands of chemically synthesized donor DNA to form a triple helix, which will then be recognized by the cell's own DNA enzyme repair systems and by the process of genetic recombination remove the mutant sequence, and insert the correct DNA sequence into the CF gene. The biochemical process takes advantage of the fact that PNA-directed triple-helix formation is known to be very effective for the sequence-specific recognition of double-helical DNA. By minimizing the chance of nonspecific off target cleavage and DNA breaks, and relying on the cell's own repair enzymes, Marie intends to avoid the risk of an immune response to the introduction of an artificial enzyme to the body. By attempting to correct the defective gene instead of replacing it with a new copy, the normal influence of regulation and control exerted by surrounding genes will also be retained. If Marie is successful, correction of the defective CF gene will be permanent and repetitive interventions should not be required, thus providing a pathway for translation to a "cure" for the vast majority of children with CF.