Researchers report oral delivery system for RNAi therapeutics

April 29 2009

Researchers at the University of Massachusetts Medical School (UMMS) report today on a novel approach to the delivery of small bits of genetic material in order to silence genes using "RNA interference"—and in the process, discovered a potent method of suppressing inflammation in mice similar to what occurs in a range of human diseases.

In the April 30, 2009 issue of the journal *Nature*, Professor Michael P. Czech, PhD, and colleagues in the Program in Molecular Medicine at UMMS describe the engineering of small encapsulating particles containing short pieces of RNA that dramatically silenced genes in mice following oral administration in small doses. The paper, "Orally delivered siRNA targeting macrophage MAP4K4 suppresses systemic inflammation," provides a possible pathway to address the most common—and daunting—challenge in the new field of RNA therapeutics: how to deliver the short strands of RNA used in gene silencing to specific tissues and cell types.

"We are very encouraged by these results, which show that oral delivery of a therapeutic dose of small, interfering RNA (siRNA) to a specific cell type in an animal model is possible, and that evidence of gene silencing using this delivery system is measurable," said Dr. Czech.

The discovery in 1998 that short strands of RNA can silence the action of a given gene changed the scientific world's understanding of how genes are regulated. Highly specific and highly potent, "RNA

interference" or "RNAi" has become both a crucial laboratory technique and widely studied for potential therapeutic applications; the explanation of the mechanism of RNAi was recognized with the 2006 Nobel Prize in Medicine, awarded to UMMS Professor Craig C. Mello, PhD, and collaborator Andrew Z. Fire, PhD, of Stanford University; since the discovery, laboratories around the world have focused on the potential of RNAi to silence genes with high specificity, low toxicity and minimal immune system response.

But how to deliver tiny strands of genetic material into cells in a living organism has been a formidable obstacle. In this paper, Czech and colleagues chose to target a particular type of cell in the immune system called a "macrophage," a type of white blood cell that engulfs and digests cellular debris and responds to invading organisms by stimulating the immune response. Because macrophages control the inflammatory response in diseases such as rheumatoid arthritis and atherosclerosis (a precursor to heart disease), they represent an attractive target for drug delivery.

To move short strands of RNA into the macrophages, the researchers exploited a distinctive characteristic of yeast particles: the ability to be engulfed and digested by macrophages. By using these yeast particles as a delivery shell, they were able to deliver siRNAs targeting a gene known for its key role in the inflammatory response—and turn it off. The macrophages carrying the RNAi moved throughout the organism as they circulated from the digestive system (where they first encountered the particles and engulfed them) with the result that over time, a large portion of the organism's macrophages exhibited gene silencing.

The method of treating yeast particles to remove components that would cause an immune response and generate oral delivery vehicles composed of "beta1,3-D glucan" was developed by UMMS research professor and paper co-author Gary R. Ostroff, PhD. The method of using glucan

particles as a drug delivery system has been tested in a number of animal models. In December 2008, the Massachusetts Life Sciences Center awarded a three-year, $750,000 cooperative research grant to UMMS and biotech startup RXi Pharmaceuticals to investigate the development of a range of orally delivered RNAi therapeutics using the glucan particle model. (RXi was co-founded by Nobel Laureate Mello, who serves on its Scientific Advisory Board, and Czech.)

In the series of experiments, the researchers were able to silence gene expression both in vitro and in vivo, in a mouse model, at a range of doses and concentrations; oral delivery of as little as 20 micrograms per kilogram of body weight of siRNA silenced a signaling protein called MAP4K4, a key player in the inflammatory response in disease processes like arthritis. (By contrast, research studies evaluating intravenous injections of siRNAs often used concentrations from 12 to 500 times higher.)

"In the future, this paper will be viewed as a landmark in the process of translating RNAi into effective new therapies for human diseases," said Terence R. Flotte, MD, dean of the school of medicine at UMMS. "It addresses one of the most fundamental problems in the field, that of delivery of the RNAi molecule to the cells affected by the disease process."

Source: University of Massachusetts Medical School (news : web)