853. Efficacy of AAV-Mediated Expression of Acid Sphingomyelinase at Correcting the Visceral and Pulmonary Manifestations of Niemann-Pick B Disease

Article *in* Molecular Therapy · May 2004

DOI: 10.1016/j.ymthe.2004.06.761

CITATION
1

READS
27

10 authors, including:

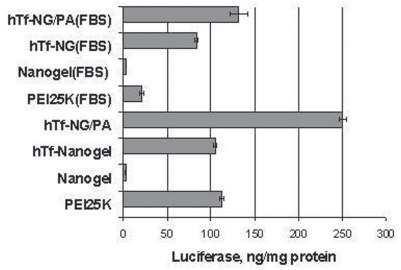
Christine M Barbon
Bluefin Biomedicine
28 PUBLICATIONS **399** CITATIONS
SEE PROFILE

Edward H Schuchman
Icahn School of Medicine at Mount Sinai
244 PUBLICATIONS **13,571** CITATIONS
SEE PROFILE

Michael Lukason
Sanofi
21 PUBLICATIONS **1,007** CITATIONS
SEE PROFILE

Scott Lonning
Bluefin Biomedicine
69 PUBLICATIONS **3,325** CITATIONS
SEE PROFILE

and effective nuclear translocation of the intact DNA. It results in high level of transfection even in presence of serum compared to the known efficient transfection agent, branched PEI25 (Figure 2). Authors believe that Nanogel can be a convenient platform for development of plasmid DNA carriers with properties of artificial virus.



INBORN ERRORS OF METABOLISM: LYSOSOMAL STORAGE AND OTHER DISEASES

853. Efficacy of AAV-Mediated Expression of Acid Sphingomyelinase at Correcting the Visceral and Pulmonary Manifestations of Niemann-Pick B Disease

C. M. Barbon,[1] R. J. Ziegler,[1] S. D. Bercury,[1] M. Cherry,[1] C. Li,[1] E. Schuchman,[2] R. J. Desnick,[2] M. J. Lukason,[1] S. Lonning,[1] S. H. Cheng.[1]
[1]Gene Transfer Research, Genzyme Corporation, Framingham, MA; [2]Mount Sinai School of Medicine, New York, NY.

Niemann-Pick B disease (NPD) is an inheritable lysosomal storage disease in which the deficient activity of the lysosomal hydrolase, acid sphingomyelinase (ASM), results in the intracellular accumulation of sphingomyelin. Accumulation is most strikingly evidenced by the presence of enlarged, often multinucleated myeloid-lineage cells in bone marrow, lung, liver, and spleen. In particular, the presence of lung infiltrates of engorged alveolar macrophages is frequently referenced in human lung biopsies and at autopsy. In NPD patients, significant morbidity is attributed to protracted pulmonary decline and recurrent respiratory infections, underscoring the importance of achieving efficacy in this organ. Systemic administration of either an AAV2/1 or 2/8 pseudotyped vector encoding human ASM under the transcriptional control of a liver-restricted promoter into acid sphingomyelinase knockout mice (ASMKO) resulted in expression of therapeutic levels of the enzyme. Transduction and subsequent expression, which was primarily hepatic-restricted, was approximately 50-100-fold higher with the AAV2/8 than with the AAV2/1 vector. Furthermore, the high levels attained using the AAV2/8 vector were readily detected in the circulation by day 4 and were sustained at the end of the 120-day study. Subsequent extrahepatic uptake of the secreted enzyme, presumably via the mannose and mannose 6-phosphate receptors, was associated with a reduction in sphingomyelin levels in all the affected tissues, including the lung. As may be expected, since the levels of expression attained with the AAV2/8 vector were significantly higher, the kinetics of sphingomyelin clearance was more rapid than with the AAV2/1 vector. Biochemical and histological analysis of ASMKO mice at days 60 and 120 post-administration demonstrated a profound reversal of lung pathology concomitant with substrate depletion to basal levels. Correction was also associated with reduced levels of the proinflammatory chemokine, MIP-1α, and normalization of the cellularity and cell differentials in the bronchoalveolar lavage fluids (BALF) of treated animals. In contrast to the severe storage inclusions and enzyme-uptake defects shown associated with alveolar macrophages from ASMKO BALF, those isolated from AAV2/8-treated mice demonstrated improved phagocytic activity and displayed morphologies that were more akin to normal macrophages. Hepatic-restricted expression of the human ASM also promoted immune tolerance to the expressed enzyme in the ASMKO mice. No antibodies to the human enzyme were detected in the animals throughout the 120-day study. Hence, AAV2/8-mediated, hepatic-restricted expression of ASM was capable of correcting the visceral storage disease and abrogating the chronic pulmonary inflammatory manifestations shown associated with NPD. This data supports the continued evaluation of the gene delivery system for the treatment of NPD.

854. Feasibility of AAV-Mediated Gene Therapy Examined Using a New Murine Model (D409V/null) of Gaucher Disease

John Marshall,[1] Kerry McEachern,[1] Jennifer B. Nietupski,[1] Julie A. Cavanagh Kyros,[1] Donna Armentano,[1] Gregory A. Grabowski,[2] Seng H. Cheng.[1]
[1]Genzyme Corporation, Framingham, MA; [2]Divisions of Human Genetics and Pathology, Cincinnati Children's Hospital Research Foundation, Cincinnati, OH.

Gaucher disease is the most common of the more than 40 currently described lysosomal storage diseases. It is caused by mutations in the gene encoding the lysosomal hydrolase, glucocerebrosidase (GC). The loss or deficiency of this enzyme results in the accumulation of the substrate, glucosylceramide (GL-1), in tissue macrophages primarily of the liver and spleen. A mouse model of the disease (D409V/null) was recently generated. Characterization of this murine model indicated that it displayed several of the biochemical and phenotypic abnormalities shown associated with the human disease. The D409V/null mice exhibited approximately only 5% of normal levels of the enzyme in the visceral tissues and consequently, elevated levels of GL-1 in the liver, spleen, lung and also the bone marrow of these animals. Associated with the abnormal storage of GL-1 in the tissues were the appearance of characteristically enlarged macrophages (Gaucher cells) similar to those observed in Gaucher patients, particularly in the liver, spleen and lung of the mice. To evaluate the potential of gene therapy for treating this disease, an AAV 2/2 and 2/8-pseudotyped vector encoding the human GC were constructed. Since secretion of GC from transduced cells occurs only in cells highly overexpressing the enzyme, efforts were made to optimize its expression. A codon optimized and CpG-reduced synthetic cDNA for human GC was placed under the transcriptional control of an α_1-antitrypsin promoter to which was appended two copies of the α_1-microglobulin enhancer (DC172). Studies showed that expression from the DC172 promoter was hepatic-restricted and was significantly higher than that attained with either a CMV or the previously described DC190 promoter (human serum albumin promoter linked to two copies of the human prothrombin enhancer). Intravenous administration of 3 X 10^{11} particles of either AAV2/2-DC172-shGC or AAV2/8-DC172-shGC into 4 weeks old D409V/null mice resulted in hepatic transduction and subsequent secretion of supraphysiological levels of GC into the systemic circulation.