3T MRI and MR spectroscopy of a Feline Model of Sandhoff Disease after AAV-Mediated Gene Therapy

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Target Audience. Studies outlined in this abstract will benefit those interested in Magnetic Resonance Imaging as an *in vivo* biomarker of neurodegenerative lysosomal storage diseases and AAV-gene therapy.

Purpose. Sandhoff disease (SD) is a form of GM2 gangliosidosis in humans that is untreatable and fatal by 5 years of age. SD is caused by mutation of the β subunit of the dimeric enzyme hexosaminidase (Hex). We performed intracranial (IC) AAV-mediated Hex gene replacement in a feline model of SD, resulting in a >four-fold increase in lifespan and marked attenuation of neurologic signs. With this profound success in IC therapy, human clinical trials are in the planning stages and development of non-invasive methods such as MRI and MRS to evaluate amelioration or disease progression are vital to predicting outcomes in humans.

Methods. Monocistronic AAV2/rh8 vectors expressing feline Hex α and β subunits (1:1 ratio) were injected bilaterally into the thalamus and deep cerebellar nuclei of Sandhoff cats (4.4×10^{12} g.c. total). MRI and MRS data were acquired on a Tesla MAGNETOM Verio scanner (Siemens Healthcare, Erlangen, Germany) at 4 months and 24 months using an 8 channel phased array wrist coil (Invivo Corp, Gainesville, FL, USA). Anatomical coronal images were acquired using 3D MPRage (magnetization-prepared rapid gradient echo) with 0.4mm isotropic resolution and TR/TE of 1900/3.3ms, followed by 2D axial T2 TSE images with TR/TE of 4630/107ms and a resolution of $(0.3x0.3x1)mm^3$, Single voxel spectroscopy (SVS) was then acquired using PRESS (Point RESolved Spectroscopy) with CHESS (CHEmical Shift Selective) water suppression, TE/TR = 30/2000ms and 32 averages. In all animals a $(4x5x13)mm^3$ voxel was placed in the parietal cortex that was well defined on the high resolution anatomical images. All metabolite peak integrals were normalized to creatine (Cr). MRI and MRS data were analyzed using EFilm 3.2 software (Merge Healthcare, Chicago) and jMRUI (Java Based Magnetic Resonance user Interface, http://www.mrui.uab.es/mrui/mrui_homePage.shtml.), respectively.

Results.

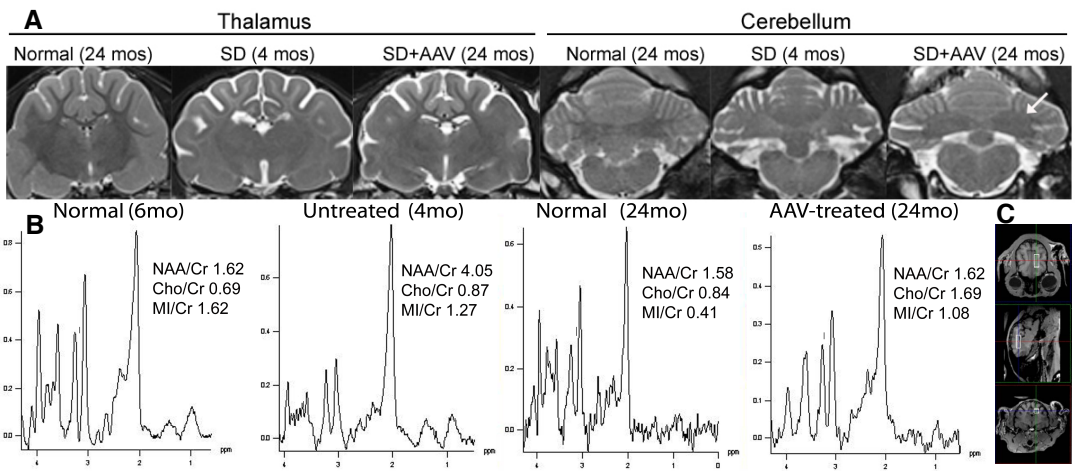

Figure 1 *3T MRI and MRS of normal and SD cat brains.* **A)** Axial T2 weighted MRIs demonstrate cortical white matter hyperintensity to gray matter at the level of the thalamus in an untreated SD cat (4 mos.). The AAV-treated SD cat has normalized white:gray matter intensities. Similarly, the deep cerebellar nuclei of the untreated SD cat are hyperintense relative to the cerebellar vermis, while DCN are hypointense in a normal cat and an AAV-treated SD cat at 24 mos of age. **B)** MRS revealed increased ratios of N-acetyl-aspartate (NAA)/Cr and Myoinositol (MI)/Cr in an untreated SD cat near the humane endpoint (4 mos.) compared to normal. At 24 mo. old, an AAV-treated SD cat showed normalized NAA/Cr, elevated choline (Cho)/Cr and MI/Cr compared to age matched control. **C)** Voxel placement was consistent between cats and located in the parietal cortex.

Discussion. Untreated SD cat brain showed white matter abnormalities and increased NAA/Cr, possibly caused by neurite production secondary to buildup of GM2 ganglioside or by decreased Cr secondary to reduced metabolism[1-3]. 3T MRI and MRS images in AAV-treated cats showed preservation of white and grey matter structures and normalization of NAA/Cr. To more closely evaluate metabolite changes, future studies are planned for determination of individual metabolite variations using an external reference solution with the phantom replacement quantification technique [4,5].

Conclusions. Gene replacement in the feline SD model results in normalization of both brain architecture and metabolites. Measurement of individual metabolites (rather than metabolite ratios) may provide a better understanding of pathogenesis.

References:

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