



## Niemann-Pick type C disease (NP-C) Gene Therapy Activation Meeting (GAM) Manoir de Clénord, Chambord, France October 18-19, 2018

The following report summarizes discussions at the NP-C Gene Therapy Activation Meeting (GAM), a first-of-its-kind satellite meeting to the bi-annual Loire-Valley Meeting (LVM) hosted by the German and Swiss Niemann-Pick family organizations on behalf of the International Niemann-Pick Disease Alliance (iNPDA). The aims of this meeting were fourfold: First, to provide expert input to the patient community on the status of gene therapy approaches in childhood-onset neurodegenerative disorders; second, to assess whether gene therapy could be a viable approach for improving NP-C; third, to align researchers from different areas and agree on a best path forward; and fourth, to discuss how to best position patients and family organizations during an NP-C gene therapy development process.

**Marie Vanier** kicked-off discussions by putting gene therapy in the context of previous and current attempts towards an NP-C therapy. She reminded the participants of some important learnings in the field since the very first clinical trials in the 1990s, e.g. that to impact the wider patient population, therapies need to modify CNS symptoms; that composite scores, albeit imperfect, have proven superior to individual disease symptoms as clinical outcome measures; that different patient subgroups respond differently to existing and probably also emerging therapies; and that under miglustat (which is approved in the EU and many other countries, but not the United States), the neurological disease continues to progress. Preliminary results of ongoing trials (HP- $\beta$ -cyclodextrin and arimocloamol) appear promising. Marie further highlighted that while in comparison to similar diseases the NP-C community has been very fortunate to have built up a good experience in conducting clinical trials, several very critical pieces – such as sufficient numbers of qualifying patients to allow multiple trials in parallel; robust quantitative biomarkers that closely track with therapeutic benefit; or experience with combination trials (although a majority of patients in ongoing trials also receive miglustat) – are still missing.

**Cristin Davidson** then outlined four key preclinical challenges to a successful NPC gene therapy, namely that due to its size NPC1 is much more difficult to integrate into current gene therapy vectors than smaller proteins; that targeting the brain and the (likely) most relevant cell types in the brain is still a considerable challenge to the field; that current gene therapy vectors at widely accepted concentrations typically transduce only a minority (<15%) of cells in a tissue; and that the expression of a foreign protein is likely to induce an immune response that may limit repetitive administrations, especially for patients who do not have any residual NPC1 protein or express only truncated versions. She then discussed the approach chosen by the team at NHGRI, using an AAV9 vector that encodes full-length NPC1 under either a CamKII or an EF1a (shortened) promoter and that is injected retro-orbitally in NPC1 knock-out mice at weaning. Impressive results both on survival as well as an improvement of a 5-item composite phenotype progression score were observed. Results were particularly encouraging when expressing NPC1 under a ubiquitous EF1-promotor using a modified AAV9-PHP.B vector. It is currently being tested whether combining gene therapy with weekly subcutaneous injections of VTS-270 will provide additional benefits.

**Michael Hughes** and **Ahad Rahim** continued discussions with an alternative approach to NP-C gene therapy in the BALB/c NPC1 knock-out mouse model. In contrast to the researchers at NHGRI, they chose to administer AAV9-hNPC1 under a neuron-specific synapsin1-promotor directly into the brain (via the cerebrospinal fluid into the lateral ventricle) and in P0 newborn mice, i.e., before the onset of any symptoms. This resulted in readily detectable NPC1 protein across multiple brain regions, including Purkinje neurons in the cerebellum. Treatment neither at higher, nor lower doses caused obvious safety signals, yet at 9-10 weeks follow-up in treated versus untreated mice, Purkinje-cell number remained considerably increased (to ~50% of wt relative to ~10% in untreated mice). Impressively, at a high dose of  $5 \times 10^{11}$  vg, Michael and Ahad's treatment regimen resulted in a significant impact on survival, with all seven treated animals now being more than one year old and still not showing obvious nervous system disease (despite emerging weight loss since peripheral disease remains untreated). Currently, combination regimes with small molecule drugs shown to decelerate NP-C disease progression are being conducted, and initial results indicate that when combined with other therapies NPC1 gene replacement may result in increased survival and reduced disease burden, even when administered at lower dosage. In summary, their results generate very promising proof-of-concept that under an optimal treatment scenario, gene therapy



will provide benefit for NP-C, and that this benefit is likely to be additive to that of approved and emerging alternative treatment modalities.

Continuing the session of NPC gene therapy preclinical studies, **Charles Vite** then talked about his experiences when translating results from mice to larger animal models. This included sharing some frustration that this translation isn't always straightforward (as he had to experience at the case of alpha-mannosidosis), yet at times can yield very promising results, as best demonstrated by videos of a highly agile dog with a gene defect in GALC that usually causes Krabbe disease. However, when AAV9-GALC was injected into the cerebello-medullary cistern at either 2 or 6 weeks of age, even at 60 weeks of age this dog model is showing only very modest signs of physical impairment, incl. near normal nerve conduction velocity and amelioration of white matter brain abnormalities. To accelerate gene therapy for NPC, Charles talked about experiments in his NPC1 cat model to explore which of six promoters tested might be the safest and enable the most desirable expression of NPC1 protein across disease-relevant brain regions. Among others, he showed encouraging results that an NPC-cat injected at 3 weeks with a modest dose of AAV9-hNPC1 into the cerebellar-medullary cistern was still able to walk at the age of 6 months, despite considerable ataxia. Higher doses and different routes of administration are currently being tested for select vectors, with experiments partially being supported by Galyatech.

Day 2 of the meeting focused on the clinical aspects of gene therapy drug development. **Marc Tardieu** provided an impressive overview of his experiences with gene therapy development for Mucopolysaccharidosis (MPS) IIIA and IIIB. Important aspects of this process were that the incidence of newly-diagnosed patients who develop these diseases “early in life” and are available for trials is only very low; that preclinical studies for these diseases had shown that long-term immunosuppression would be needed to avoid an immune- or inflammatory response to substitution of the missing NAGLU protein; and that extensive natural history studies, incl. the development of tools to quantitatively monitor the development and decline of cognition in these conditions was required prior to any clinical trial. After several years of developing the protocol, a Phase I/II clinical trial with AAV2-NAGLU in four newly diagnosed MPSIIIB patients of less than 5 years of age was initiated. This trial was initially sponsored by Pasteur Institute with the vector provided by the start-up UniQure (who later deprioritized further development of this program, despite encouraging outcomes). rAAV2-hNAGLU vector was injected at a single point in time via intra-cerebral delivery through 16 microcatheters placed in eight holes in the skull. Over an observation period of 31 months, six severe adverse events were observed that were related to the intervention due to very short hospitalization (diarrhea and benign injection). However, after an overall four years follow-up the therapy was tolerated well in all four participants. All four patients showed a slower decline in cognition than expected from natural history studies, and one of the patients does still not shown signs of brain atrophy (while brain atrophy in the three others was reduced when compared to historic controls). In all patients, NAGLU enzymatic activity could be reconstituted to ~15% of normal levels, and two of the four patients still show elevated TNFa in NAGLU-stimulated CD4 and CD8 T-cells (under continued immunosuppression). Thus, overall, this trial showed promising efficacy at moderate safety risks and has set the stage for discussions how to best continue MPSIIIB gene therapy development in a larger Phase 3 trial, as long as an industrial sponsor could be found.

**Heiko Runz** then contributed at the case of Spinal Muscular Atrophy (SMA) to what are important considerations at later stages of drug development. Heiko mentioned that among others drug development for SMA benefited from the considerably higher prevalence of this disease, despite still being considered a rare disease; the existence of a single main genetic defect common to almost all SMA patients and a fairly well-understood biological mechanism of disease; and the ability and framework to diagnose patients early in the course of their disease. Heiko further showed that success of the very first registrational Phase 2/3 trial for SMA with a drug that is based on an antisense-oligonucleotide (ASO) modality was followed by several more refined trials that then also included SMA patients who had not met inclusion criteria of the initial trial. Approval of the drug (which is now marketed under the name Spinraza), and the observation that treatment is most efficacious the earlier patients are receiving the drug, led to the successful addition of SMA to newborn screening panels in several countries. Despite its promising efficacy and safety profile, a very high pricing structure for Spinraza makes it difficult for SMA patients in many healthcare systems to get access to the drug. Also, there are currently 16 known and promising competitor programs, incl. four based on gene therapy, that - despite the high prize - challenge continued returns-on-investment for Biogen (as the manufacturer of Spinraza), an issue that may also become relevant for



other rare diseases once the first successful therapies are emerging (Disclaimer: Heiko Runz is a full-time employee at Biogen).

The final session of the meeting consisted of a **general discussion** on gene therapy for NP-C. The discussion centered around five questions:

### **1. Is gene therapy at all a viable approach for NP-C and should it be pursued?**

All attendees agreed that recent progress in the field has increased the general optimism that NP-C could indeed be amenable to gene therapy, although it is certainly not a “low-hanging fruit” example for this still new modality. However, despite such general optimism and based on the still modest examples of successful gene therapies for much “easier” diseases, attendees also agreed that many unanswered questions remain before an NPC1 gene therapy can confidently be tested in patients, and that rushing into first-in-human trials (as promoted already now by several start-up companies) could be rather detrimental to success. Nevertheless, the time was considered right to start discussions on how a potential first gene therapy trial for NP-C might look like, especially since protocol development takes time and before the background of emerging alternative NP-C therapies.

### **2. What is still missing with respect to preclinical development?**

There was consensus that AAV9 is currently the most promising vector for gene therapies targeting the brain, although it is still open whether a wildtype or modified AAV9 will emerge as the best delivery system. Also, commonly used promoters are well-enough understood, yet a decision will need to be made whether a gene therapy should address preferentially/exclusively neurons, or how much effort should be spent on targeting also other cell types and tissues. Unclear (among others) are still the optimal viral dose used for an initial first-in-human trial; what is the optimal administration route; to what extent does NPC1 gene replacement trigger an immune response; whether NPC1-II1061T mice respond similarly to gene therapies as the currently tested NPC1 knock-out mice; how well findings in mice translate to larger model organisms; and whether promising results from animal studies will at all be transferable to humans (which has not been the case for several other diseases, with positive and negative examples existing).

### **3. How should clinical development best be approached?**

The discussion to this question centered around which checkboxes need to be ticked to decide that the time is right to move forward and start with experiments in patients, i.e. when do we feel ready to initiate a clinical trial? Attendees agreed that in the gene therapy field there’s a whole lot that we just don’t understand, yet for other diseases this has not hindered researchers to test their approach in small numbers of patients. Ideally, it would be an expert group to define the criteria needed for moving forward (such as which defined percentage of Purkinje cells should successfully express NPC1 in NP-C cats; or which safety data, improvement in survival and motor function, and if possible cognition, suffice, ideally as well in the cat model). As for previous NP-C trials, late-infantile or early juvenile patients were again considered to be the likely most informative patient group for a clinical trial, although due to the lack of alternative therapeutic options and because discussions with regulators might be easier, future NP-C trials are likely to pay more attention to adult patients.

### **4. How can and should patients be involved?**

It was generally recognized that commercial aspects will play a tremendous factor in the decision at what point to start a clinical trial, and while there is an in-principle desire among researchers as well as among families to collaborate and align consensually behind one common way forward, realities may stand against this (as the field has experienced already during previous NP-C trials). Attendees agreed that patients do have a strong say in driving future developments since they are the ones who control their data and their participation. Also, while the iNPDA should not deviate from its status as an “honest broker”, this organization has the most influential voice and should be encouraged to live up to its expectations to inform, educate and guide families on the current status and options available to patients. It was remarked, that what is treatment success may need to be defined individually for each family, and that a very important factor will be whether NP-C gene therapy improves overall quality of life.

### 5. How can we best operationalize the next steps forward?

All attendees agreed that it would be detrimental to future progress if patients are being “locked up” in (too early) clinical trials that are based on mediocre science. It is important to hold the academic-driven momentum in the field for as long as possible, and to eventually align behind a single most promising approach before initiating a race towards being the first one in clinical trials. Communication – between competing groups, towards the industry and regulators, and towards and among the families – will be a most critical element to ensure success, and the major groups in the field should get together to inform each other on their respective status and ideally distribute tasks so that unnecessary duplication of efforts can be avoided. A consensus statement of what are the minimal requirements for kickstarting and successfully running gene therapy trials for NP-C were considered as very desirable joint output and a very concrete next step that the attendees (ideally jointly with other groups) could take on. All agreed that the GAM was an excellent first step towards such collaborative approach, and we hope that similar meetings will arise at a regular basis to allow continued exchange, and to rapidly yet thoroughly deliver gene therapy as a successful and causative treatment for NP-C.

[Summary drafted by Heiko Runz]



**GAM participants** (from left to right):

Christoph Poincilit, Michael Hughes, Marc Tardieu, Marie Vanier, Marc Patterson, Heiko Runz, Charles Vite, Cristin Davidson, Ahad Rahim, Ning Lü.