1/14/24

Re: Scientific justification for CIRM support for n-of-1 gene therapy clinical trial

The field of medicine is rapidly evolving through the 'omics' revolution. The past 20 years has seen reports of thousands of disorders previously without clear causes now solved through identification of genetic causes. These conditions affect nearly every field of medicine and age range, and cut across socioeconomic and racial barriers. Most conditions are without proven therapy. This represents a huge unmet medical need. This also represents an opportunity to advance the scientific pursuit of disease mechanisms and novel therapeutics.

In 2017 the results from the clinical trial of the antisense oligonucleotide (ASO) nusinersen demonstrated potent effects on survival in the otherwise lethal neonatal spinal muscular atrophy [1]. ASOs are usually designed to treat a mutation rather than the disease itself [2,3]. ASOs represent a powerful drug platform, but because of exquisitely specific binding to RNA nucleotides, most potential disease-altering ASOs could only be used in small patient populations, and would thus likely never be profitable for drug companies [4,5]. Yet many patient advocacy groups now recognize the potential for these programmable drugs to correct protein expression, offering hope for new cures.

Clinical trials, and n-of-1 clinical trials in particular, can offer cluses into cellular disease mechanisms, and can be viewed as scientific experiments. Some critical questions are: 1] Which diseases will demonstrate clinical benefit by an ASO? 2] Can we exploit this information to understand how correction of protein expression leads to improved outcome? In the case of nusinersen, the dramatic clinical response changed the thinking about disease mechanisms and plasticity. With CIRM's existing investment in IPSCs models of disease, there is exciting potential to advance novel drug platforms like ASOs towards unmet medical need. For instance, the Gleeson Lab at UCSD is a recipient of a CIRM DISC2 grant that, in collaboration with n-Lorem, tests preclinical efficacy of ASOs to correct differential gene expression signatures in patient IPSC cell models of pediatric brain disease prior to treatment.

A fair question is whether n-of-1 clinical trials can be viewed as scientific experiments or comparable to standard clinical trials. Our view is that they can be viewed as scientific experiments because: 1] They are established with quantitative endpoints, using fit-for-purpose outcome measures, measured against the patient's prior natural history. These include outcomes 'measured by machine', which should have little potential for placebo effects. 2] All data including drug properties, outcome measures, and side effects are entered into the public domain, and thus can be leveraged by others to expand the work. 3] Meta-analysis of multiple patients with the same disease or gene or ASO will further evidence clinical benefit. And while standard clinical trials are concerned with differences in the population *means*, the only way an n-of-1 clinical trial will prove positive is if the outcome is so substantially different from the patient's history as to convince even skeptics this would not be possible without targeted intervention.

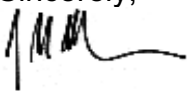
ASOs can deliver clinical benefit by silencing toxic gain-of-function mutations; correcting splicing defects to restore protein production; or targeting unique genetic vulnerabilities such as micro-RNAs, upstream open reading frames or naturally occurring

Joseph G. Gleeson, MD
Department of Neurosciences and Pediatrics
UCSD School of Medicine
Chief Medical Officer, n-Lorem Foundation

Laboratory for Pediatric Brain Disease
Rady Endowed Chair in Neuroscience
Rady Children's Institute for Genomic Medicine
9500 Gilman Dr., La Jolla CA 92037

antisense transcripts. n-Lorem and efforts like it, have developed workflows to first assess the mutation and clinical status, and then simultaneously develop safe and potent ASOs as well as suitable outcome measures to assess clinical benefit in an FDA approved n-of-1 clinical trial. The outcomes of these trials will fill a critical void in medical and scientific knowledge, and have the potential to usher in a wave of novel therapeutics to leverage the 'omics' breakthroughs in medicine.

Sincerely,



Joseph G. Gleeson, MD

References

1. Finkel, R.S., Mercuri, E., Darras, B.T., Connolly, A.M., Kuntz, N.L., Kirschner, J., Chiriboga, C.A., Saito, K., Servais, L., Tizzano, E., et al. (2017). Nusinersen versus Sham Control in Infantile-Onset Spinal Muscular Atrophy. N Engl J Med *377*, 1723–1732. 10.1056/NEJMoa1702752.
2. Crooke, S.T., Liang, X.-H., Crooke, R.M., Baker, B.F., and Geary, R.S. (2021). Antisense drug discovery and development technology considered in a pharmacological context. Biochem Pharmacol *189*, 114196. 10.1016/j.bcp.2020.114196.
3. Crooke, S.T., Witztum, J.L., Bennett, C.F., and Baker, B.F. (2018). RNA-Targeted Therapeutics. Cell Metab *27*, 714–739. 10.1016/j.cmet.2018.03.004.
4. Gleeson, J.G., Bennett, C.F., Carroll, J.B., Cole, T., Douville, J., Glass, S., Tekendo-Ngongang, C., Williford, A.C., and Crooke, S.T. (2023). Personalized antisense oligonucleotides "for free, for life" - the n-Lorem Foundation. Nat Med *29*, 1302–1303. 10.1038/s41591-023-02335-2.
5. Mittal, S., Tang, I., and Gleeson, J.G. (2022). Evaluating human mutation databases for "treatability" using patient-customized therapy. Med *3*, 740–759. 10.1016/j.medj.2022.08.006.