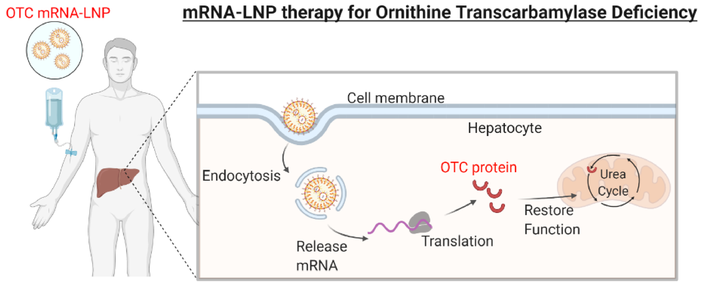
25

Preclinical Evaluation of Modified mRNA for the Treatment of OTC Deficiency

Daly, O.,[1] Lam, K.,[1] Meffen, T.,[1] Reid, S.,[1] Yaworski, E.,[1] Tyler, S.,[1] Vlatkovic, I.,[2] Mahiny, A.J.,[2] Reinholz, J.,[2] Besold , K.,[2] Fesser, S.,[2] Lepper, M.,[2] Berte, N.,[2] Lindemann, C.,[2] Marlot, P.T.,[2] Kuhn, A.N.,[2] Karikó, K.,[2] Lutwyche, P.,[1] Esau, C.,[1]

[1] Genevant Sciences Corporation, 155-887 Great Northern Way, Vancouver, BC V5T 4T5. [2] BioNTech RNA Pharmaceuticals GmbH, An der Goldgrube 12, 55131 Mainz, Germany

ABSTRACT



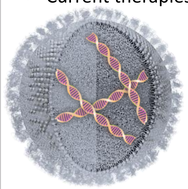
Ornithine transcarbamylase (OTC) deficiency is a rare X-linked genetic disorder characterized by complete or partial lack of the OTC enzyme. OTC plays a key role in the urea cycle, and its absence leads to inability to metabolize ammonia and is associated with permanent brain damage and death. Genevant and BioNTech are collaborating to co-develop an mRNA therapy enabled by Genevant's industry-leading nucleic acid delivery capabilities to treat OTC deficiency.

Codon-optimized, nucleoside-modified mRNA encoding human OTC (hOTC) was encapsulated in lipid nanoparticles and administered to both wild-type (WT) and OTC-deficient mice (OTC spf^{ash}). OTC spf^{ash} mice are an excellent model for study of OTC deficiency, with residual OTC expression levels of ~5% compared to WT and a low tolerance to a high protein diet. Here, we challenged OTC spf^{ash} mice with this diet to demonstrate the effects of our OTC mRNA-LNP in a diseased state. We subsequently tested the lead mRNA-LNP in a multi-dose NHP study, observing robust OTC protein expression and no changes in liver safety signals at clinically relevant dose levels.

INTRODUCTION

OTCD is a devastating disease

- 1/56,500 incidence with no ethnicity or geography spared.
- Most patients present with symptoms such as developmental delay, liver damage
- Current therapies are limited (e.g., liver transplant, ammonia scavengers)



Opportunity for mRNA as therapy

- mRNA therapeutics would allow patients to make the missing or defective enzyme inside the cell to correct the disease-causing deficiency
- Effective delivery of mRNA into hepatocytes and the ability to repeat dose are necessary

RESULTS

Assessment of hOTC mRNA variants in Mice

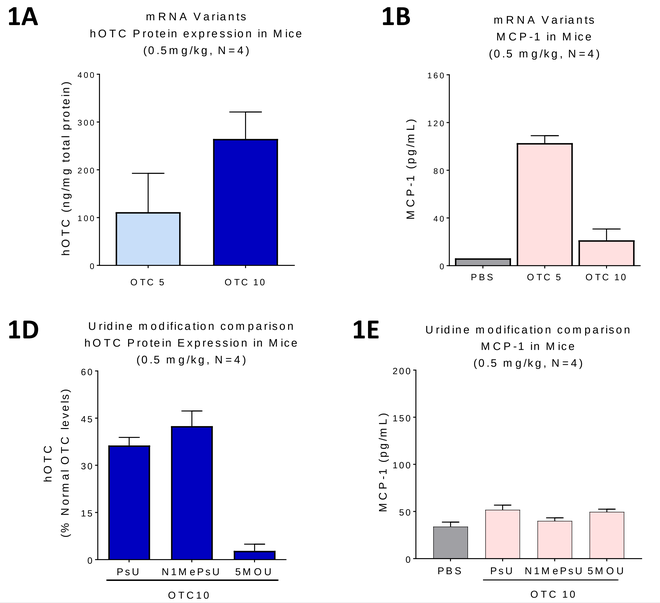


Figure 1. mRNA design affected expression and immune stimulation. OTC 10 outperformed other potential mRNA candidates, demonstrating high hOTC expression **(A)** and low MCP-1 induction (**B**). The low immune stimulation was maintained even at high dose levels **(C).** OTC 10 mRNA containing either pseudouridine (PsU) or m1-pseudouridine (N1MePsU) translated better with less immune activation than mRNA containing 5-methoxyuridine (5MOU) **(D&E).**

Testing hOTC mRNA-LNP in NHP

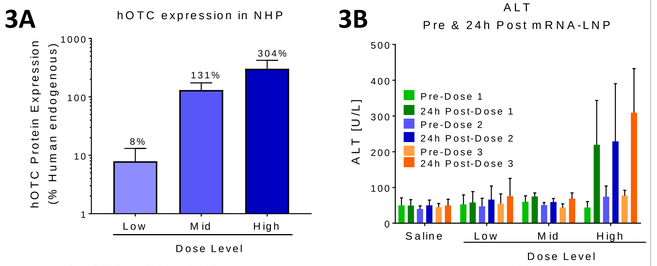


Figure 3. OTC mRNA-LNP safety and protein expression in NHP. To assess clinically relevant dose levels, animals were treated weekly (x3) with hOTC encoding mRNA-LNP. Robust expression was observed in liver samples analyzed by mass spectrometry 24h post 3rd dose **(A)**. Liver parameters such as ALT showed no change at low dose levels with only modest increases at higher dosages **(B).**

Survival benefit of hOTC mRNA-LNP in a disease model

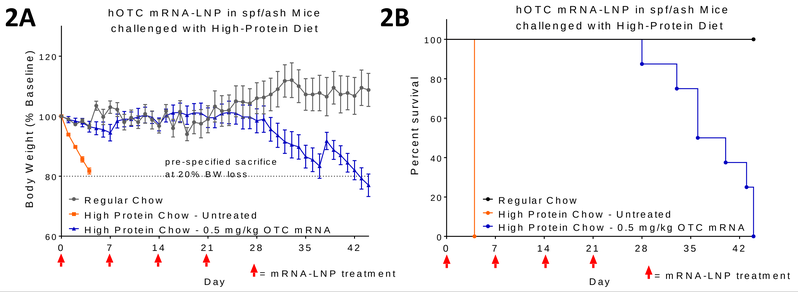


Figure 2. hOTC mRNA-LNP improved survival of OTC-spf^{ash} mice, a model of human OTCD. Four weekly doses of human OTC mRNA-LNP protected spf^{ash} mice from a High-Protein challenge thus demonstrating a restoration of the urea cycle **(A).** Body weight changes also highlight the benefit of increasing hOTC expression in the challenge model **(B).**

CONCLUSIONS

- This mRNA-LNP product candidate showed robust protein expression and a favorable safety profile in rodents and NHP
- In a well accepted murine model of OTCD, weekly doses of OTC mRNA-LNP increased survival significantly, using bodyweight loss as a surrogate endpoint. Many animals survived well after the final dose.
- In NHP, three weekly doses of hOTC-encoding mRNA-LNP resulted in robust protein expression and no changes in liver parameters at low dose levels.
- Taken together, these data demonstrate a potential OTC mRNA-LNP therapy suitable for clinical development.

CONTACT INFORMATION

Owen Daly: owen.daly@genevant.com , telephone: 778-650-4210
Genevant Sciences Corporation
155 - 887 Great Northern Way
Vancouver, BC V5T 4T5