

Title: Development of Novel Class of Therapeutic Oligonucleotides Based on Small Molecule Screening

Julia Alterman, Hong Cao, Marie Didiot, Mehran Nikan, Matthew Hassler, Anastasia Khvorova
RNA Therapeutics Institute, Program in Molecular Medicine, Small Molecule Screening Core,
Department of Biochemistry & Molecular Pharmacology, University of Massachusetts Medical
School

Anastasia.Khvorova@umassmed.edu

Abstract:

Highly inefficient transit of oligonucleotides from outside cells to the intracellular compartments where functional activity of oligonucleotides takes place is the most serious limitation to the practical realization of a full potential of oligonucleotide-based therapies. Several classes of oligonucleotide therapeutics (ONT), including antisense oligonucleotides (ASO), hydrophobically modified siRNAs (hsiRNA), GalNAc-conjugated siRNAs, and LNP-formulated siRNAs have validated biological efficacy and are in clinic [1, 2]. In all cases, the fraction of oligonucleotides reaching the intended place of biological function is surprisingly low, with the majority of molecules being trapped in wrong cellular compartments, resulting in low efficiency and clinically limiting toxicity [3].

We have recently completed a cell-based screen using the LOPAC library and identified a panel of small molecules that alter cellular localization and dramatically enhance the efficacy of hydrophobically modified siRNAs (hsiRNAs) developed previously [4] (Navaroli et al 2013). In the presence of top two hits (Guanabenz and Phenamil), we have observed a dose-dependent enhancement of oligonucleotide efficacy, with both a significant increase in cellular uptake and decrease in EC50 values.

Use of small molecules as enhancers and modulators of oligonucleotide therapeutic efficacy is a new paradigm in formulation development with wide implications on compounds in clinic and future developments.

1. de Fougerolles, A.R., *Delivery vehicles for small interfering RNA in vivo*. Hum Gene Ther, 2008. **19**(2): p. 125-32.
2. Vaishnav, A.K., et al., *A status report on RNAi therapeutics*. Silence. **1**(1): p. 14.
3. Stalder, L., et al., *The rough endoplasmic reticulum is a central nucleation site of siRNA-mediated RNA silencing*. EMBO J, 2013. **32**(8): p. 1115-27.
4. Byrne, M., et al., *Novel Hydrophobically Modified Asymmetric RNAi Compounds (sd-rxRNA) Demonstrate Robust Efficacy in the Eye*. J Ocul Pharmacol Ther, 2013.