SAT424

Impact of ESC+ Technology on the Hepatic Safety Profile of GalNAc-Delivered, HBV-Targeted RNAi Therapeutics

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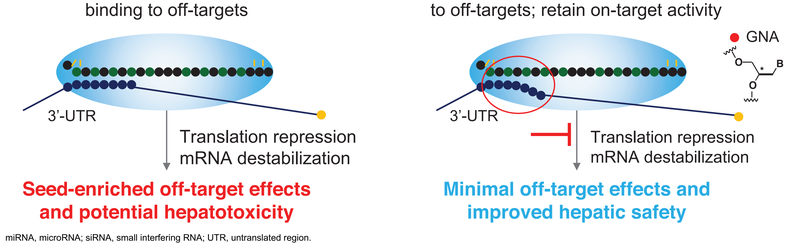
Introduction

- RNA interference (RNAi) therapies have the potential, alone or in combination with other therapies, to achieve a functional cure in patients with chronic hepatitis B virus (HBV) infection[1]
- The hepatic safety of RNAi therapies is important in the treatment of chronic HBV infection, as this represents a patient population with underlying liver disease
- ALN-HBV and VIR-2218 are investigational HBV RNAi therapeutics designed to target all HBV transcripts
- The sequences of VIR-2218 and ALN-HBV are identical; however, VIR-2218 incorporates Enhanced Stabilization Chemistry Plus (ESC+) technology, which incorporates glycol nucleic acid (GNA) within the seed region
- ESC+ technology is designed to reduce off-target seed-mediated binding while maintaining on-target activity[2,3]
- This is hypothesized to result in an improved hepatic safety profile



miRNA, microRNA; siRNA, small interfering RNA; UTR, untranslated region.

Objectives

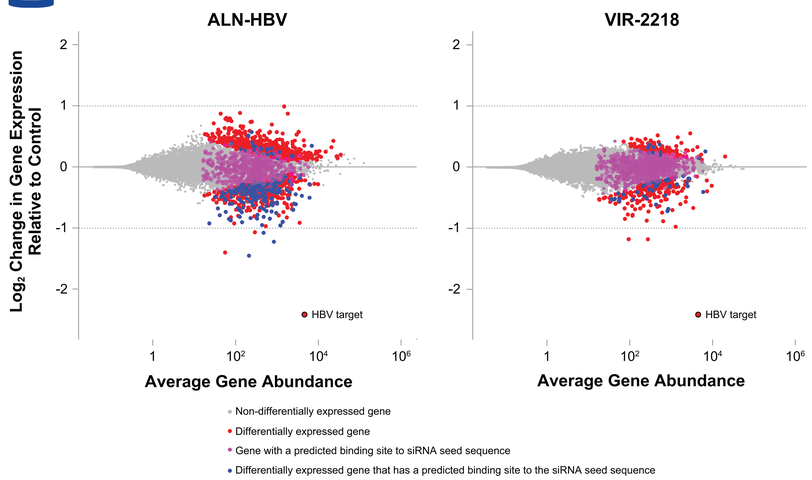
- To compare off-target silencing and hepatic safety profile of VIR-2218 and ALN-HBV *in vitro, in vivo,* and in healthy human volunteers

Methods

- To compare the *in vitro* specificity of VIR-2218 vs ALN-HBV, RNA-sequencing (RNAseq) analysis was used to evaluate differential gene expression in HepG2.2.15 cells treated with VIR-2218 or ALN-HBV
- To compare the *in vivo* hepatic safety of VIR-2218 vs ALN-HBV, human-alanine aminotransferase-1 (h-ALT1) levels were evaluated in chimeric mice with humanized livers following administration of VIR-2218 or ALN-HBV
- To determine the hepatic safety profile of VIR-2218 or ALN-HBV in healthy volunteers, changes in liver function tests after administration of single doses of ALN-HBV, up to 3 mg/kg (Study ALN-HBV-001; ClinicalTrials.gov NCT02826018) or single doses of VIR-2218, up to approximately 15 mg/kg (Study VIR-2218-1001; NCT03672188), were evaluated

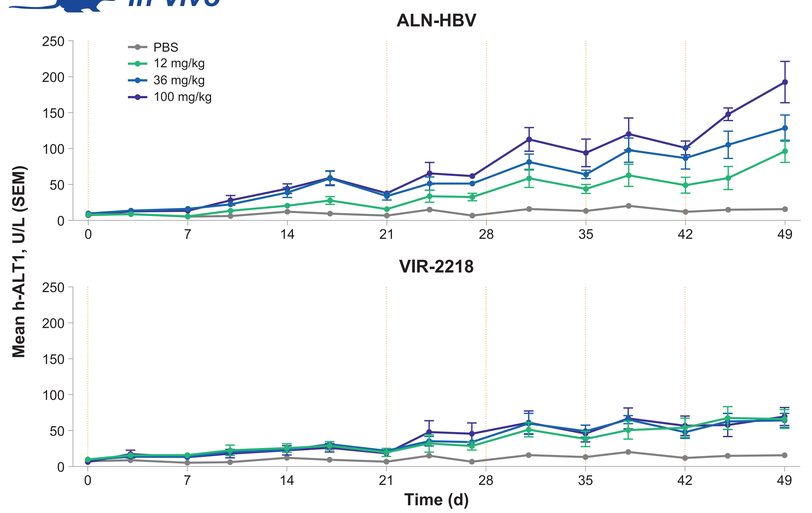
Results

In Vitro



- RNA seq analysis in HepG2.2.15 cells showed fewer differentially expressed genes and a lower magnitude of gene dysregulation, supporting reduced off-target effects with VIR-2218 compared with ALN-HBV

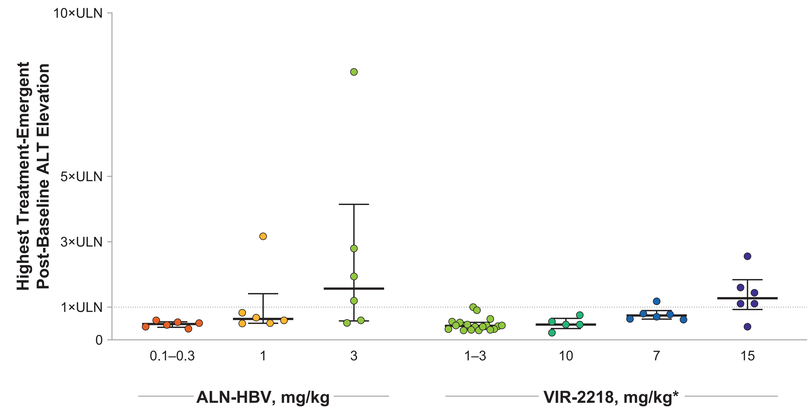
In Vivo



Dotted lines indicate dose timing. PBS, phosphate-buffered serum; SEM, standard error of mean.

- h-ALT1 levels were markedly lower following administration of VIR-2218 compared with ALN-HBV at equivalent dose levels up to 100 mg/kg
- A dose-dependent increase in h-ALT1 levels was observed with ALN-HBV, but not VIR-2218

Human: Treatment-Emergent Post-Baseline ALT Elevations in Healthy Volunteers With Normal ALT at Baseline



*Approximate mg/kg dose based on an average adult weight of 60 kg; fixed doses ranged from 50–900 mg. ULN, upper limit of normal.

- No post-baseline ALT elevations to >ULN in the VIR-2218 or ALN-HBV cohorts were associated with increases in bilirubin >ULN
- No changes in functional status of the liver (eg, albumin, coagulation parameters) or clinical signs/symptoms of hepatic dysfunction were observed in any ALN-HBV- or VIR-2218-treated patient

Conclusions

- Relative to ALN-HBV, the ESC+ siRNA VIR-2218 was shown to have:
  - Improved *in vitro* specificity by reducing seed-mediated off-target effects
  - Substantially decreased propensity to cause ALT elevations in a humanized liver chimeric mouse model
  - Substantially decreased propensity to cause ALT elevations in healthy volunteers at dose levels anticipated to be clinically relevant
- Evaluation of the safety and antiviral activity of VIR-2218 in patients with chronic HBV infection is presented in abstract AS068; extended follow-up is ongoing

References: 1. Flisiak R, et al. Expert Opin Biol Ther 2018;18:609-17; 2. Nair J, et al. J Am Chem Soc 2014;136:16958-61; 3. Foster D, et al. Nucleic Acids Res 2018; 4. Janas MM, et al. Nature Commun 2018;9:723; 5. Schlegel MK, et al. J Am Chem Soc 2017;139:8537-46.

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