Pre-Clinical Profiling of a Novel Class of Orally Bioavailable Small Molecules Potently Inhibiting Hepatitis B and D Virus Entry

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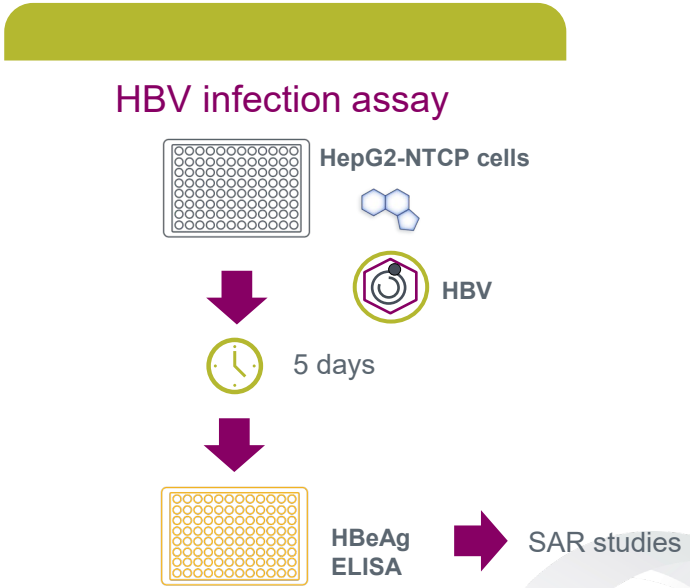
Presenter Disclosures

- Marc P Windisch is an employee and stockholder of Assembly Biosciences, Inc.

There is a Need for Orally-Bioavailable HDV Drugs

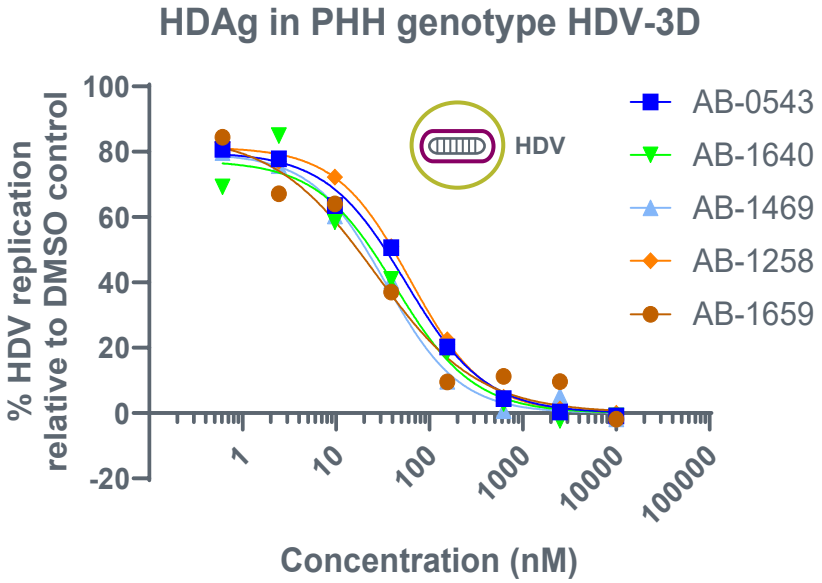
- HDV is a satellite virus that requires the HBV envelope to infect hepatocytes [1,2]
  - approx. 12 million chronically infected patients, worldwide [3]
- HDV infection is the most severe form of viral hepatitis.
  - incidence rates of HCC are higher in HDV-positive vs negative individuals [4]
- Limited treatment options for HDV
  - IFNα → weekly injections [5]
  - Bulevirtide (BLV), only approved drug (EMA) [6,7] → daily injections

Screening for HBV/HDV entry inhibitors targeting NTCP



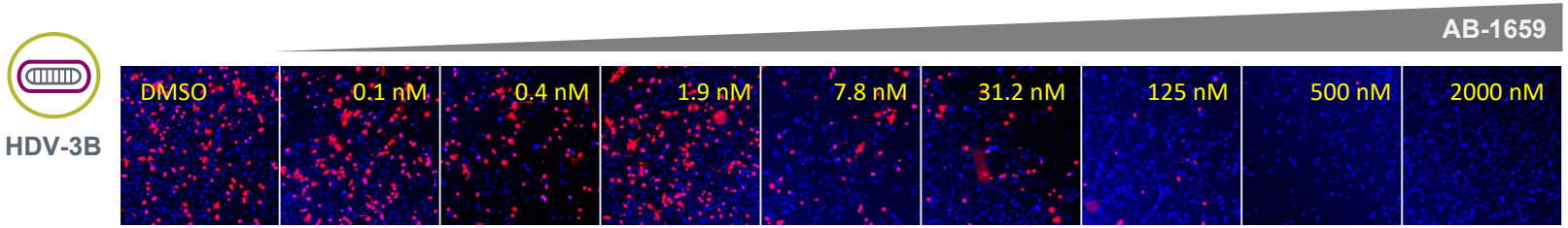
1) Hughes S, et al. *Lancet*. 2011;278:73-85. **2)** Urban S, et al. *Gut*. 2021;70:1782-94. **3)** Stockdale A, et al. *J Hepatol*. 2020;73(3):523-32. **4)** Kushner T, et al. ***J Hepatol.*** **2015;63:586-92**. **5)** Pegasys. Prescribing information. Hoffmann-La Roche, Inc. c/o Genentech, Inc; 2021. **6)** Kang C, et al. *Drugs*. 2020;80:1601-5. **7)** European Medicines Agency. Hepcludex. https://www.ema.europa.eu/en/medicines/human/EPAR/hepcludex. Accessed September 11, 2023.

Small-Molecule Entry Inhibitors Efficiently Block HDV Infection in PHHs

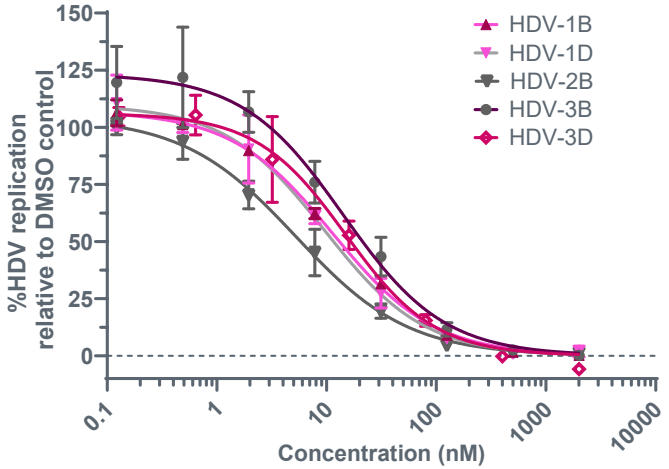


Compound	HDV EC_{50} (nM) genotype 3D *HDAg in-cell ELISA*
AB-0543	62
AB-1640	45
AB-1469	45
AB-1258	45
AB-1659	**29**

AB-1659 Potently Inhibits Multiple HDV Genotypes Enveloped With HBV Genotype B or D in HepG2-NTCP Cells



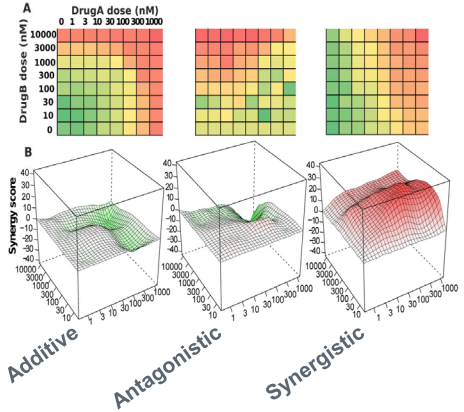
HDAg is shown in red and cell nuclei are shown in blue



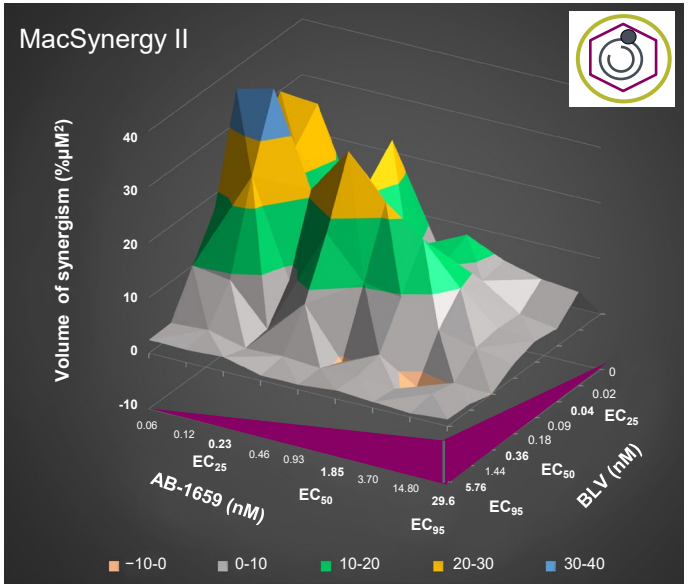
HDV/HBV genotypes	HDV EC_{50} (nM) HDAg
1B	11
1D	10
2B	5
3B	14
3D	15

Proof-of-Concept Study: AB-1659 Has Synergistic Antiviral Activity in Combination With BLV in HBV-Infected HepG2-NTCP Cells

Overview of drug combination analyses[1]



Three-dimensional model of analyzing AB-1659 & BLV drug-drug combinations

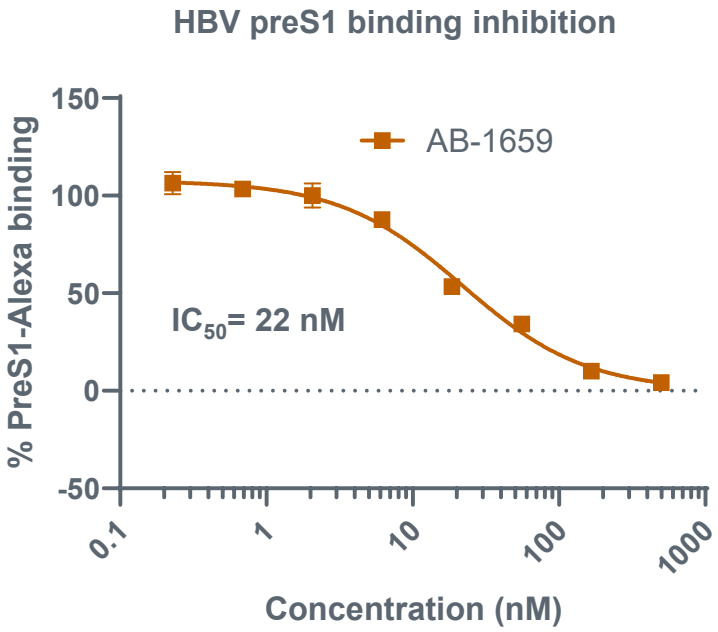


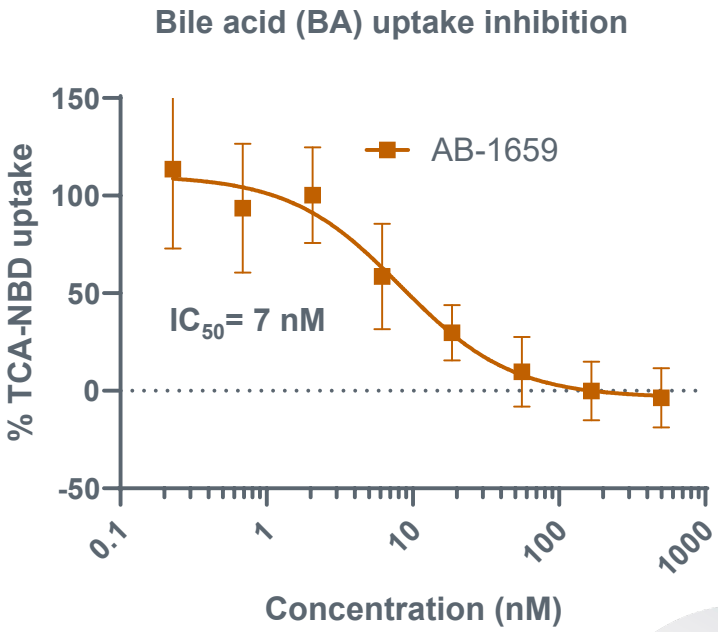
Volume of synergism=590
Volume of antagonism=−2.8

Volume of synergism (%μM²)	Log volume	Interpretation
<25	<2	Insignificant
25-50	2-5	Minor synergism
50-100	5-9	Moderate synergism
100-1000	>9	Strong synergism

1) He L, et al. *Methods Mol Biol*. 2018;1711:351-98.

AB-1659 Inhibits HBV PreS1 Binding and NTCP-Dependent Bile Acid Uptake in HEK293 Cells Demonstrating That NTCP is The Target



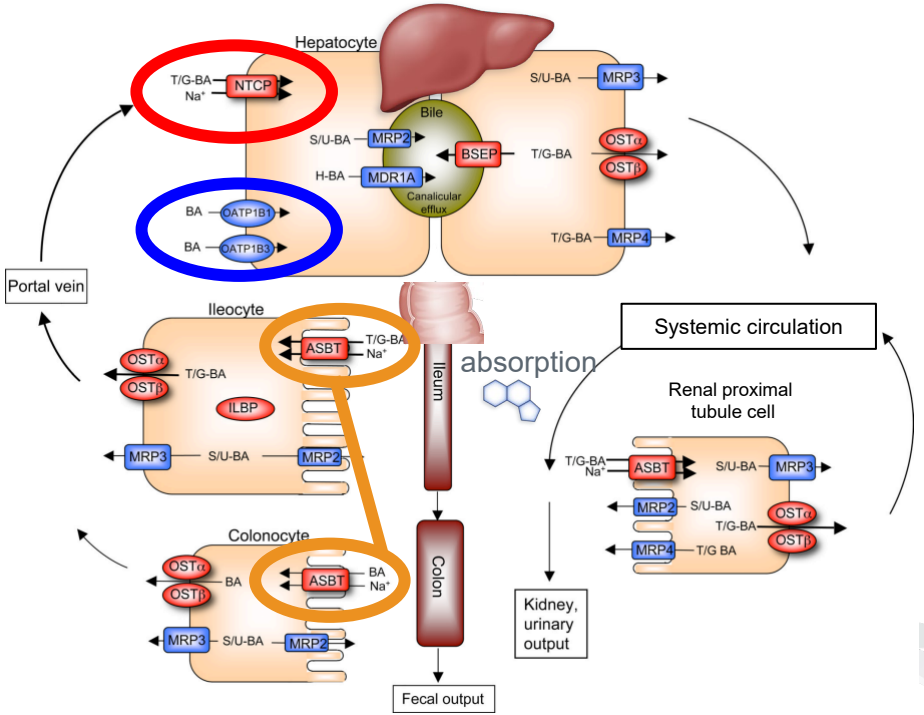


TCA, taurocholic acid

AB-1659 Is a Selective NTCP Inhibitor

Transporter selectivity (expressed as IC_{50} BA transporter / IC_{50} NTCP)	
NTCP	0.007 μM
OATP1B1	342
OATP1B3	41
ASBT	219
BSEP	5205
OATP1A2	2740

Complex interplay of BA transporter for BA homeostasis[1]

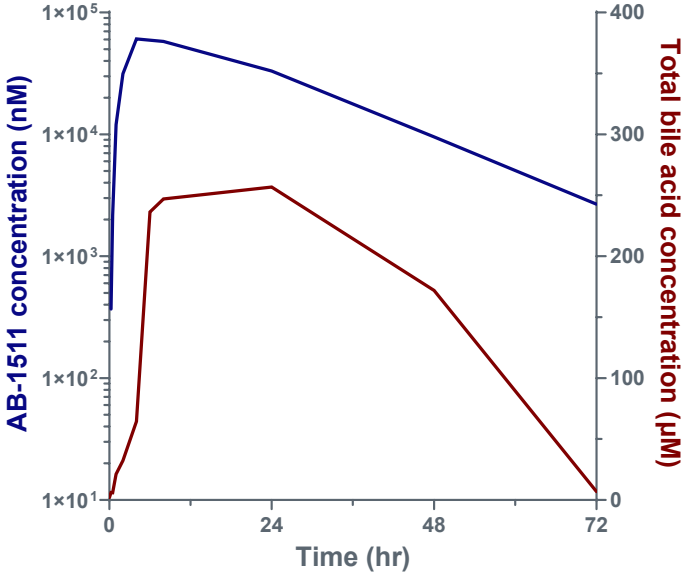


1) Adapted from Dawson P, et al. *Handb Exp Pharmacol*. 2011;201:169-203.

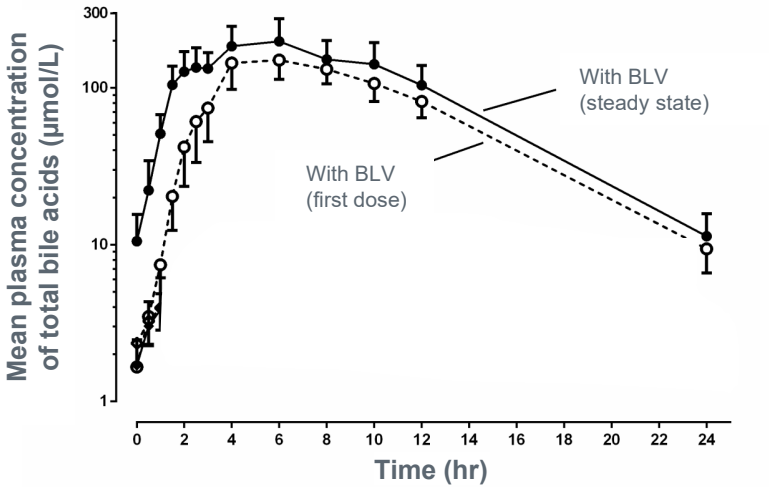
ASBT, apical sodium-dependent bile acid co-transporter; OATP, organic anion transporting polypeptide; BSEP, bile salt export pump

AB-1659 Has a Favorable PK Profile and Leads to Transient Total Bile Acid Elevations Demonstrating Target Engagement in NHPs

Pharmacokinetic/Pharmacodynamic (PK/PD) study in plasma
30 mg/kg oral single dose of AB-1659 in NHPs



Plasma concentration-time profiles of total bile acids during therapy with BLV



Adapted from Blank A, et al. *Clin Pharmacol Ther.* 2018;103:341-48.

NHPs, non-human primates

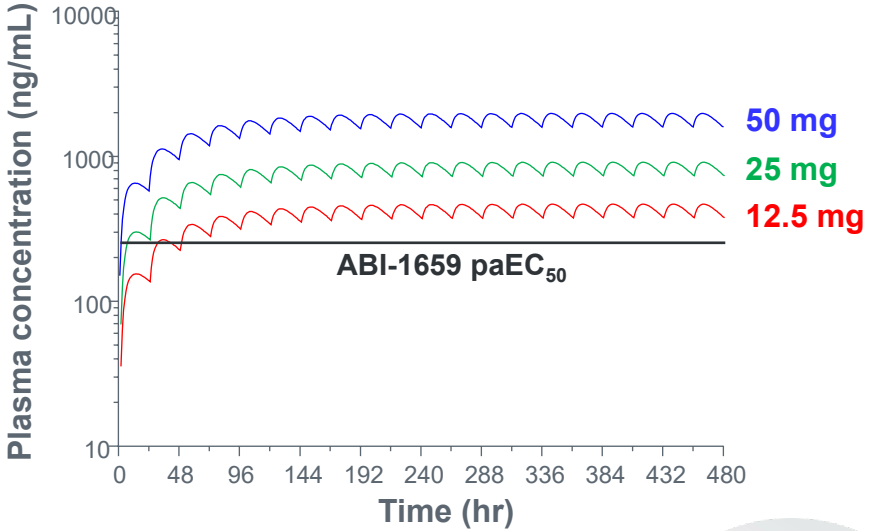
AB-1659 Has a Favorable ADME Profile That Is Consistent With Once-Daily Treatment

AB-1659 ADME profile

Properties	
$t_{1/2}$, hr	36
Bioavailability (F%)	99
hERG inhibition (10 µM)	1.2%
Liver microsome (LM) human/NHP/rat/mouse % remaining at 45 min	94 / 100 / 100 / 82
Ames test	negative
Protein-adjusted (pa)HDV EC_{50}	~500 nM

hERG, human ether-a-go-go-related gene

Human dose projection



Conclusions

- A novel class of highly-potent, orally-bioavailable HBV/HDV entry inhibitors was identified with favorable drug-like properties

- AB-1659 was selected for further characterization and demonstrated:
  - Potent activity against multiple HBV and HDV genotypes
  - Synergistic antiviral activity when combined with BLV
  - Selective inhibition of NTCP vs other human bile acid transporters
  - A favorable PK/PD profile in non-human primates
  - Projected human dose of ≤50 mg orally once daily

- We anticipate nominating a development candidate from this program in 2023

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- This study was sponsored by Assembly Biosciences, Inc.