Second-generation hepatitis B virus core inhibitors ABI-H2158 and ABI-H3733 have enhanced potency and target coverage for both antiviral inhibition and covalently closed circular DNA establishment activities

Dawei Cai, Marc Evanchik, Ran Yan, Renuka Kumar, Yuhua Zong, Qi Huang, Richard Colonno, Kathryn Kitrinos, Luisa M Stamm, William Delaney

Assembly Biosciences, Inc., South San Francisco, CA, USA

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Disclosures

- William Delaney is an employee and stockholder of Assembly Biosciences, Inc.

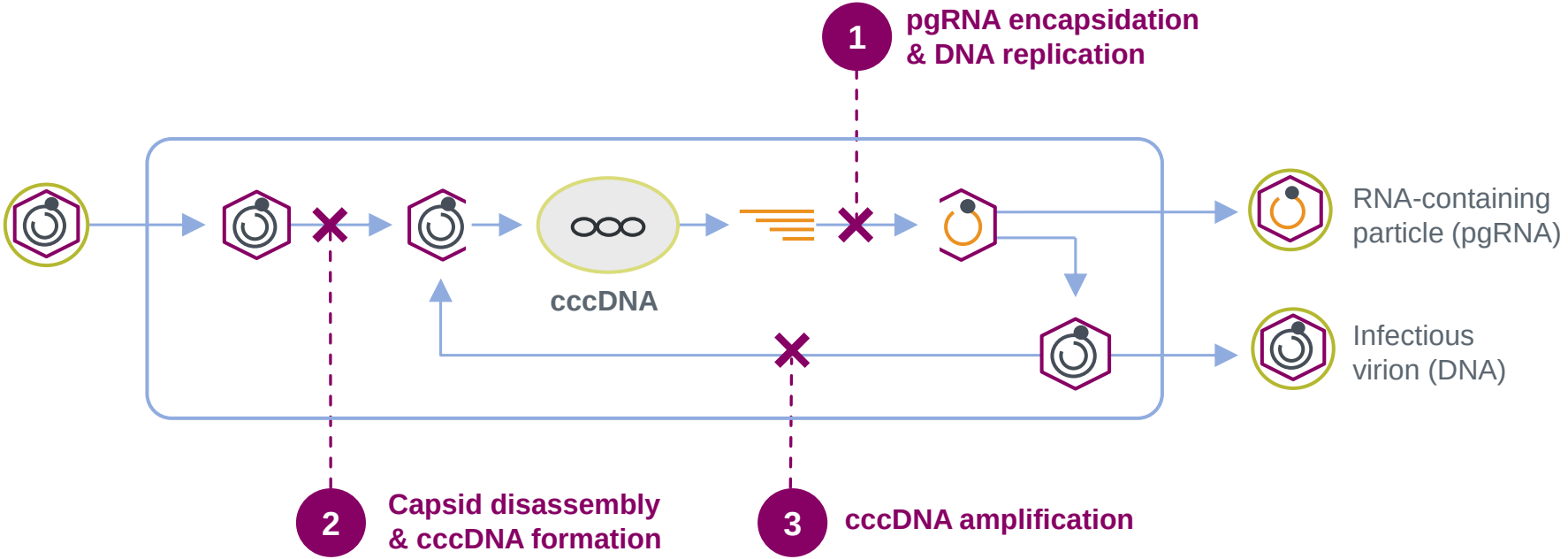
Introduction

- Core inhibitors are a new class of HBV antivirals with the potential to improve treatment of cHBV patients
- Core inhibitors interfere with multiple steps in the HBV replication cycle and result in enhanced antiviral suppression when used with NrtIs
- Assembly has three structurally-distinct core inhibitors in clinical development[1–3]
  - Vebicorvir: Being tested in two Phase 2 triple-combination studies
  - ABI-H2158: Being tested in a Phase 2 study in combination with NrtIs
  - ABI-H3733: Recently completed Phase 1a PK study in healthy adults

1. Huang Q, et al. *Antimicrob Agents Chemother*. 2020;AAC.01463-20. **2.** Huang Q, et al. *Hepatol*. 2017;66(1 Suppl): 493A. **3.** Huang Q, et al. EASL 2019. Oral Presentation.

cHBV, chronic hepatitis B virus; HBV, hepatitis B virus; NrtI, nucleos(t)ide analog reverse transcriptase inhibitor; PK, pharmacokinetic.

Core Inhibitors Target Multiple Steps of the HBV Replication Cycle



- Core inhibitors are more potent against viral replication (1) vs cccDNA formation (2, 3)
- Potent activity against both replication **and** cccDNA formation may be crucial for optimal patient responses

cccDNA, covalently closed circular DNA; HBV hepatitis B virus; pgRNA, pregenomic RNA.

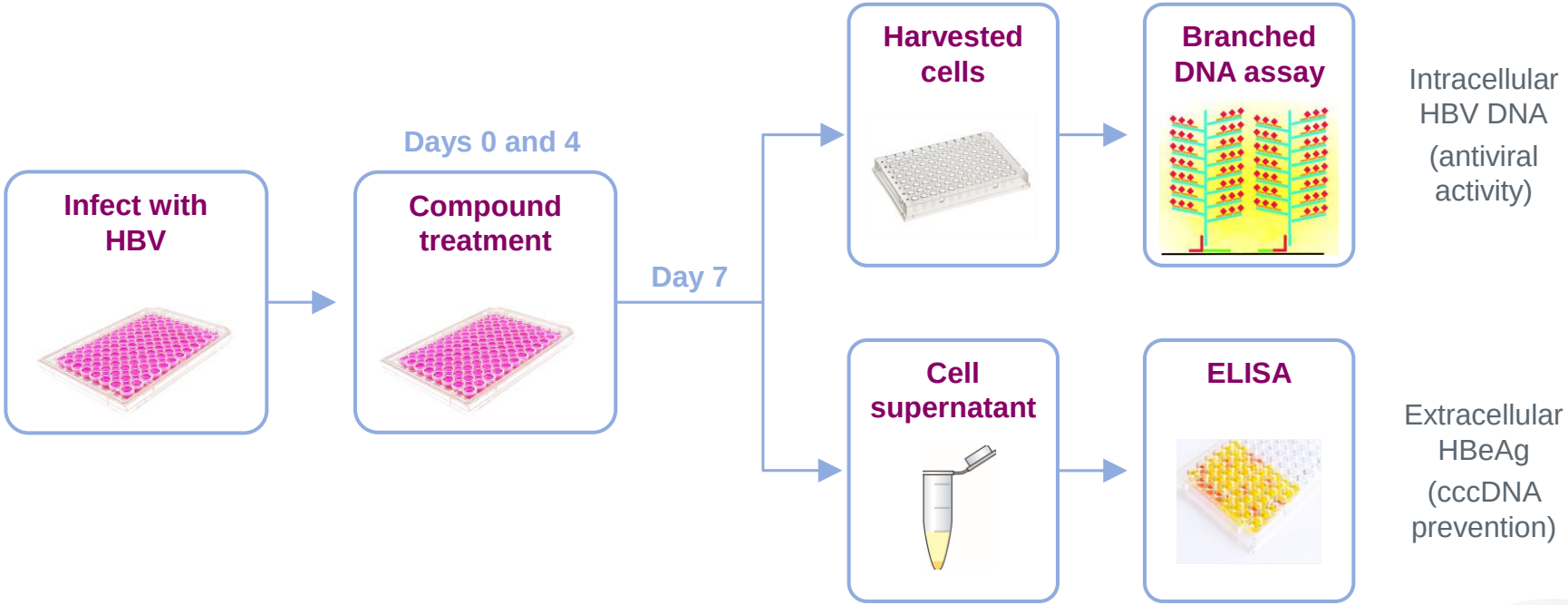
Objective

- To evaluate human plasma and estimated liver concentrations of VBR, 2158, and 3733 relative to their respective protein-adjusted EC_{50} values for
  - Assembly and release of new viral particles (**antiviral activity**)
  - Prevention of formation of new cccDNA (**cccDNA prevention**)

The clinical efficacy of antivirals depends not only on intrinsic potency but also on protein binding and the drug exposure achieved

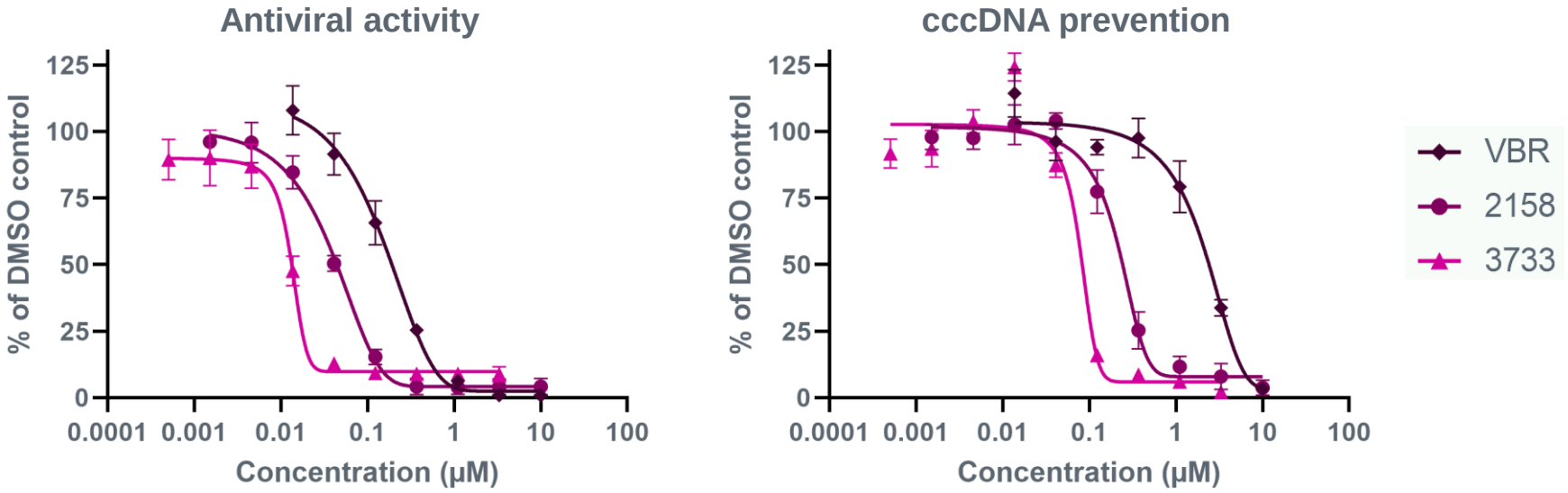
cccDNA, covalently closed circular DNA; EC_{50}, half maximal effective concentration; VBR, vebicorvir.

Determining EC_{50} in Primary Human Hepatocytes



cccDNA, covalently closed circular DNA; EC_{50}, half maximal effective concentration; ELISA, enzyme-linked immunosorbent assay; HBeAg, hepatitis B "e" antigen; HBV, hepatitis B virus.

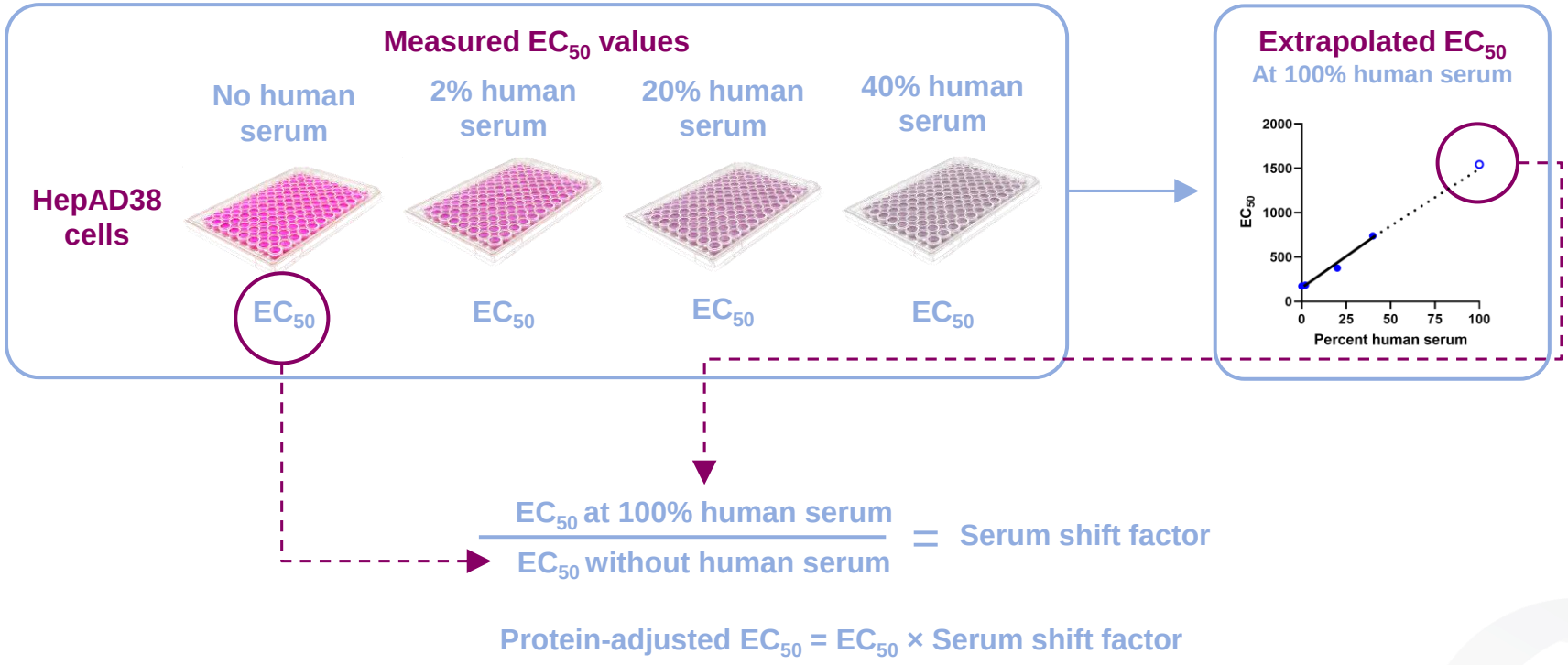
Potency of VBR, 2158, and 3733 in PHH



	VBR (n = 6)	2158 (n = 2)	3733 (n = 3)
Antiviral activity, EC_{50} (nM)	154 ± 18	41 ± 2.6	12.4 ± 2.7
cccDNA prevention, EC_{50} (nM)	2210 ± 310	204 ± 38	61.7 ± 9.8

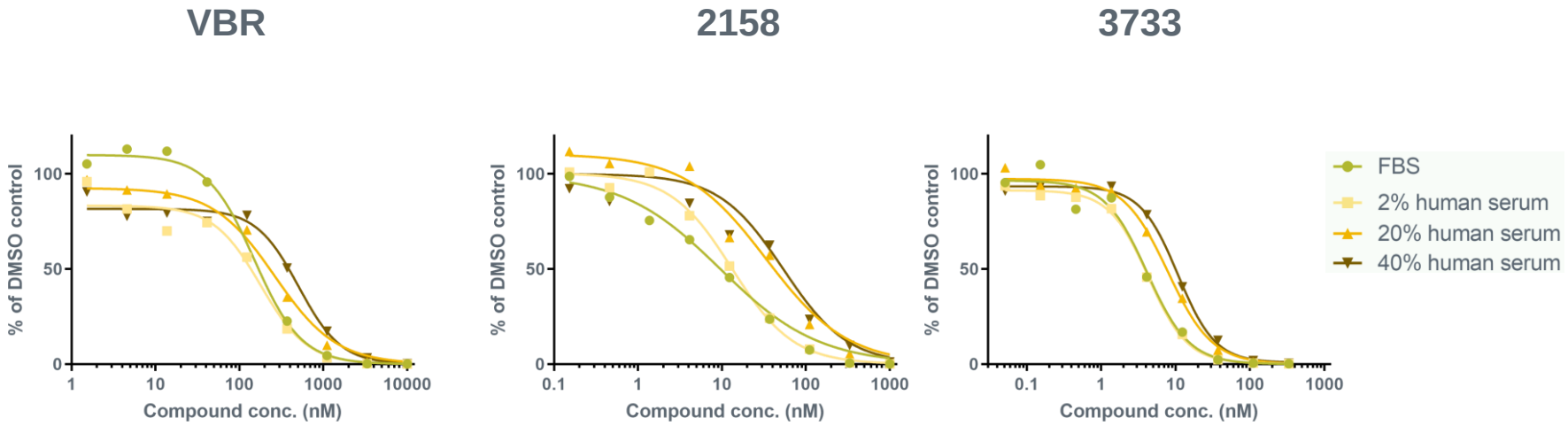
cccDNA, covalently closed circular DNA; DMSO, dimethyl sulfoxide; EC_{50}, half maximal effective concentration; PHH, primary human hepatocyte; VBR, vebicorvir.

Assay to Measure Effect of Human Serum on EC_{50} (Serum Shift Assay)



EC_{50}, half maximal effective concentration.

Serum Shift Values for VBR, 2158, and 3733



Compound	VBR (n = 4)	2158 (n = 4)	3733 (n = 4)
Serum Shift Factor	7.6 ± 1.1	7.5 ± 1.5	7.2 ± 1.9

DMSO, dimethyl sulfoxide; FBS, fetal bovine serum; VBR, vebicorvir.

PK Values Determined From Phase 1 Studies

- Pharmacokinetic values, including steady state minimum concentration (C_{min}; 300 mg QD dosing), were calculated from Phase 1 studies
  - VBR: Study ABI-H0731-101 (cHBV patients, NCT02908191)[1]
  - 2158: Study ABI-H2158-101 (cHBV patients, NCT03714152)[2]
  - 3733: Study ABI-H3733-101 (healthy adults, NCT04271592)[3]

PK parameter	VBR 300 mg QD	2158 300 mg QD	3733 300 mg QD
C_{min} (nM)	3081	5852	2454
$T_{1/2}$ (h)	24	19	25
C_{max} (nM)	8536	18146	5609
AUC (h·ng/mL)	22700	112700	40020

1. Yuen et al. *Lancet Gastroenterol Hepatol*. 2020;5:152-66; **2**. Agarwal et al. EASL 2020 poster presentation; **3**. Data on file.
AUC, area under the curve; cHBV, chronic hepatitis B; C_{max}, steady state peak concentration; C_{min}, steady state minimum concentration; PK, pharmacokinetic; QD, once daily; $T_{1/2}$, half-life; VBR, vebicorvir.

Plasma C_{min} and Protein-Adjusted EC_{50} Comparisons

Mechanism	Plasma parameter	VBR 300 mg QD	2158 300 mg QD	3733 300 mg QD
Antiviral activity	C_{min}/EC_{50}	20	143	204
	$C_{min}/paEC_{50}$	3	19	28
cccDNA prevention	C_{min}/EC_{50}	1	29	40
	$C_{min}/paEC_{50}$	0.2	3.8	5.5

- After 300 mg QD doses in humans, all core inhibitors achieved trough (minimum) plasma concentrations several fold above their antiviral activity protein-adjusted EC_{50}s ($paEC_{50}$)
- 2158 and 3733 achieved trough plasma concentrations several fold above their cccDNA prevention $paEC_{50}$

cccDNA, covalently closed circular DNA; C_{min}, steady state minimum concentration; EC_{50}, half maximal effective concentration; pa, protein-adjusted; QD, once daily; VBR, vebicorvir.

Liver C_{min} and Protein-Adjusted EC_{50} Comparisons

Mechanism	Liver Parameter	VBR 300 mg QD	2158 300 mg QD	3733 300 mg QD
	Liver:plasma ratio	18	5	6
Antiviral activity	C_{min}/EC_{50}	360	714	1227
	$C_{min}/paEC_{50}$	47	95	170
cccDNA prevention	C_{min}/EC_{50}	25	143	237
	$C_{min}/paEC_{50}$	3	19	33

- Estimated liver concentrations for VBR, 2158, and 3733 were several fold above both antiviral activity and cccDNA prevention $paEC_{50}$ s

cccDNA, covalently closed circular DNA; C_{min}, steady state minimum concentration; EC_{50}, half maximal effective concentration; pa, protein-adjusted; QD, once daily; VBR, vebicorvir.

Conclusions

- Core inhibitors are more potent against the formation of new virions (antiviral activity) vs generation of new cccDNA; we believe both activities are likely to be important for maximal impact in patients
- Plasma C_{min} values for VBR, 2158, and 3733 at safe and well-tolerated doses are significantly above protein-adjusted EC_{50} values for antiviral activity
- 2158 and 3733 have plasma C_{min} values significantly above protein-adjusted EC_{50} values for cccDNA prevention
- Liver C_{min} values for VBR, 2158, and 3733 are predicted to be significantly above the protein-adjusted EC_{50} for both antiviral activity and cccDNA prevention
- Assembly has a fourth core inhibitor program moving into development in mid-2021
  - Targeted plasma C_{min} concentrations are >25-fold above protein-adjusted EC_{50} for cccDNA

cccDNA, covalently closed circular DNA; C_{min}, steady state minimum concentration; EC_{50}, half maximal effective concentration; VBR, vebicorvir.

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