ABSTRACT OS-2478: Oral Presentation

VRON-0200, a therapeutic HBV vaccine with an intrinsic checkpoint inhibitor, elicits broad CD8$^+$ T cell responses and sustained antiviral declines in preclinical studies

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Disclosures

- **Dr. Ertl** is a co-founder of Virion Therapeutics, LLC & Professor, Vaccine and Immunotherapy Center, The Wistar Institute*
- **Dr. Luber** is a co-founder of Virion Therapeutics, LLC and has equity shares in the company
- **Mr. Magowan** has equity shares in Virion Therapeutics, LLC

Virion Therapeutics has provided funding to the Ertl laboratory at The Wistar Institute

*Dr. Ertl is an inventor of patented technologies licensed to Virion, she receives research funding support from and has an equity interest in Virion Therapeutics and consults for several companies.

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Background
CD8+ T Cell Impairment and Chronic HBV Infection

- **CD8+ T cells become impaired during chronic HBV infections, resulting in loss of viral control**
- **Immune modulators & therapeutic vaccines for chronic HBV have shown limited clinical benefits[1–5]**
  - "Rescue" of T cells with PD-1 checkpoint blockade is limited by:
    - ✓ Irreversible epigenetic changes in most HBV-specific T cells
    - ✓ Serious "off target" side effects in an otherwise healthy population
- **Stimulation of naïve HBV-specific T cells by traditional therapeutic vaccines**
  - Likely ineffective as most T cells to immunodominant HBV-specific epitopes are already activated
  - Optimized vaccine approaches that induce a response of naïve T cells to *de novo* epitopes may be able to restore viral control

CD, cluster of differentiation; HBV, hepatitis B virus; PD-1, programmed cell death protein 1.

1. Boni C, et al. *Gastroenterology.* 2019;157:227-41; 2. Gane E, et al. *J Hepatol.* 2019;71:900-7; 3. Janssen H, et al. AASLD. 2020. Abstract 0829; 4. Zoulim F, et al. *Hum Vaccin Immunother.* 2020;16:388-399; 5. Jansen D, et al. *Clin Trans Immunology.* 2021;10:e1232. doi: 10.1002/cti2.1232.

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VRON-0200

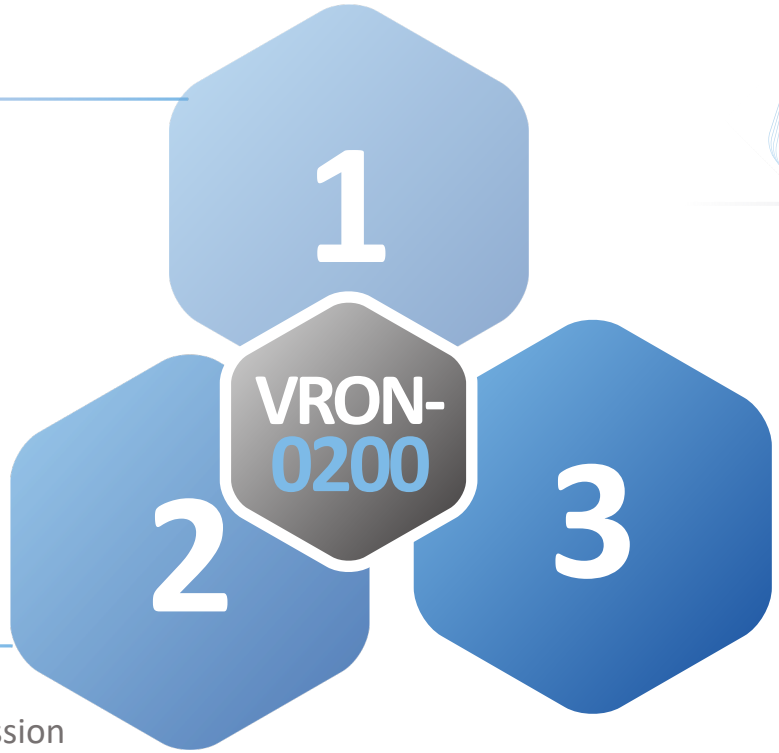
HBV Core & Pol Targeted Therapeutic Vaccine

1. Vaccine antigen:
HBV core & pol antigens

- Consensus sequences of core & pol for genotypes A–D
- S antigen, an immune decoy, was excluded
- Optimized for MHC class I binding epitopes using HLA prediction algorithms
- Regions associated with preventing flares/viral breakthrough included[1–4]

2. Vaccine carrier:
Chimpanzee-derived adenoviral vectors

- Limited pre-existing neutralizing antibodies – optimal transgene product expression
- Heterologous vectors with no cross immunity – prime and boost evaluations



3. Vaccine adjuvant:
Glycoprotein D

- Genetically encoded inhibitor of an early T cell checkpoint
- Strengthens and broadens $CD8^+$ T cell activation
- Locally acting, rapidly cleared; low risk for "off target" AEs

Challenge model: AAV8-1.3HBV

Liver trophic AAV resulting in high loads of HBV in serum

AEs, adverse events; HLA, human leukocyte antigen; MHC, major histocompatibility complex; pol, polymerase.

1. Hoogeveen RC, et al. *Gut.* 2019; 68:893-904; 2. Rivino L, et al. *J Clin Invest.* 2018;128:668-81; 3. Cheng Y, et al. *Sci Immunol.* 2019;4:eaau6905; 4. Boni C, et al. *Gastroentrology*. 2012;143:963-73.

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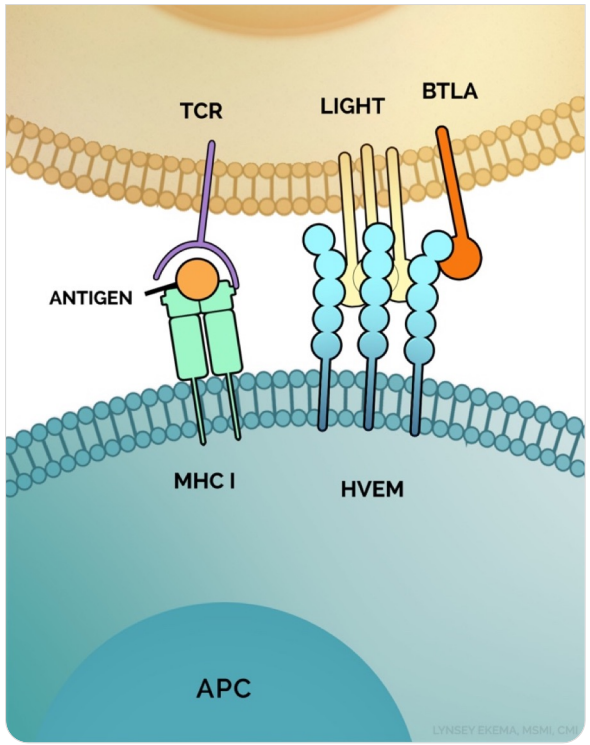
Herpes Simplex Virus Glycoprotein D

The Genetically Encoded Checkpoint Inhibitor Adjuvant in VRON-0200

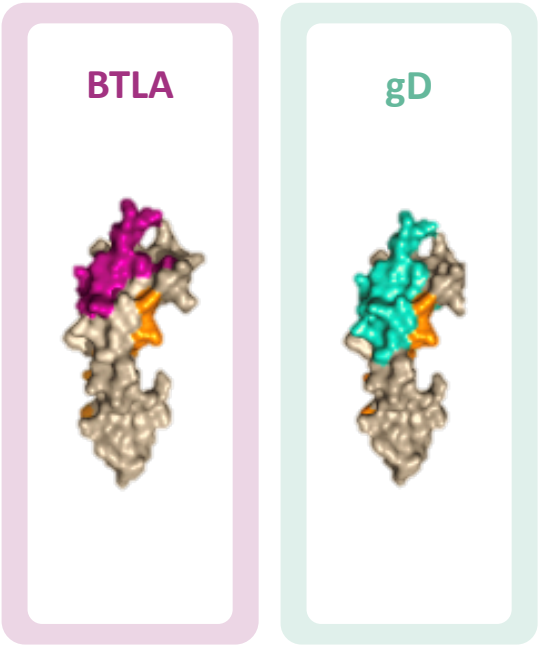
HVEM Complex in Regulating T cell Activation



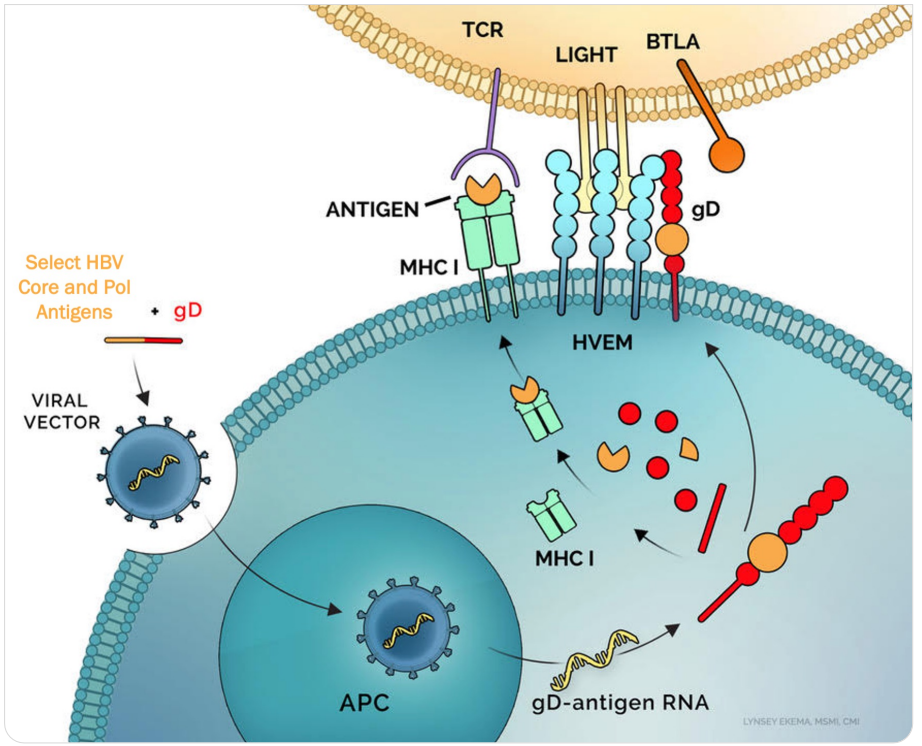
BTLA-HVEM – inhibition
LIGHT-HVEM – stimulation
BTLA-HVEM-LIGHT - inhibition

gD & BTLA Share HVEM Binding Site

HVEM crystal structure[1]



gD BTLA-HVEM Blockade Enhances and Broadens T cell Activation



APC, antigen presenting cell; BTLA, B- and T-lymphocyte attenuator; HVEM, herpes virus entry mediator; pol, polymerase; TCR, T cell receptor.

1. Stiles KM, et al. *J Virol.* 2010;84:11646-60.

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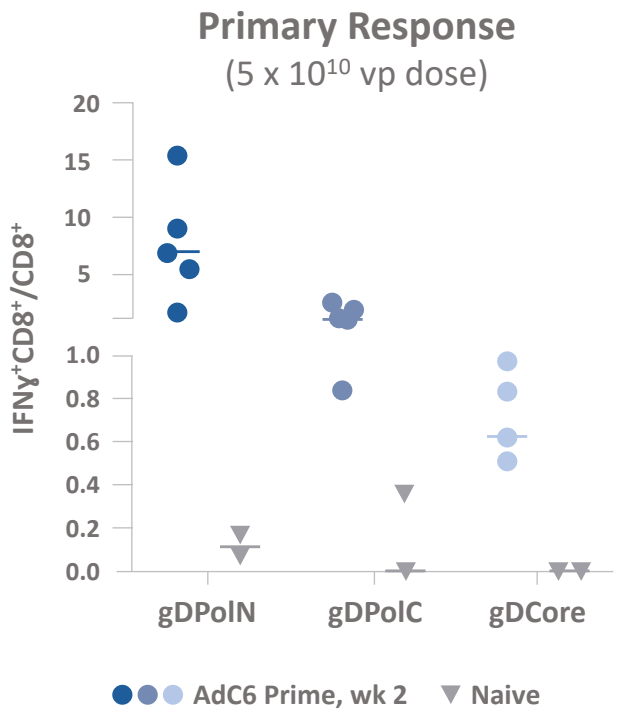
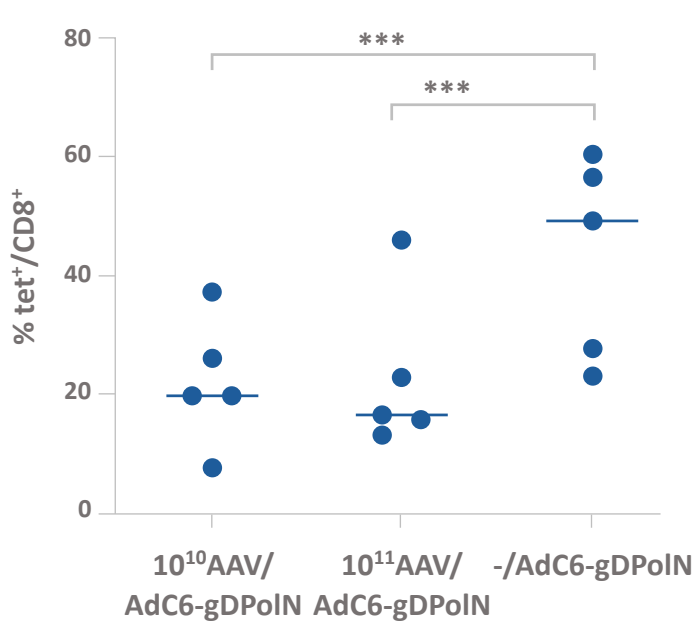
Initial Preclinical Proof of Concept
Single Antigen Studies

VIRION THERAPEUTICS

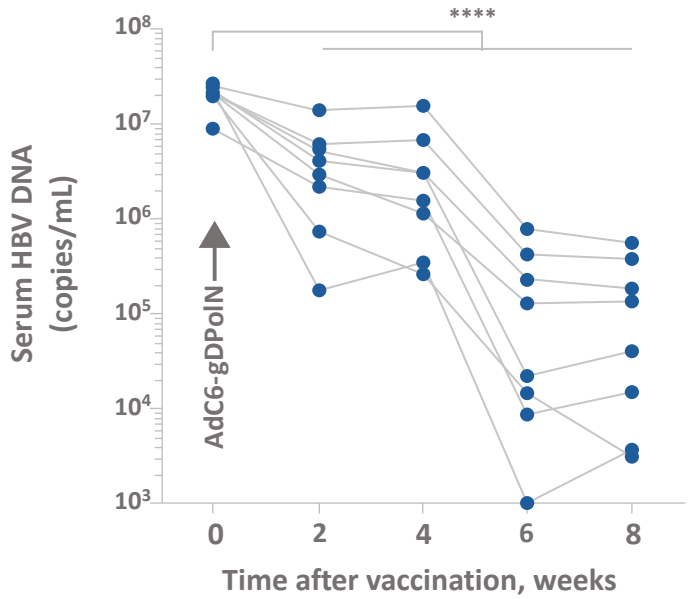
1 CD8+ T Cell Responses to All 3 Antigens



2 Vaccine-Induced T Cells Traffic to the Liver (gDPolN)



3 Multi-Log Antiviral Declines with Single IM Injection (AAV; gDPolN)



P-value between 0.0001–0.001 ;*P-value <0.0001 by ordinary one-way ANOVA.
All experiments performed with C57Bl/6 mice.
AAV, adeno-associated virus; AdC, chimpanzee derived adenovirus vector; ANOVA, analysis of variance; IFNγ, interferon gamma; IM, intramuscular; vp, virus particles.

1. Hasanpourghadi M, et al. *J Hepatol.* 2020;73(Suppl 1):S572 Presented at the EASL/DISL Meeting 2020 (Abstract 1303); 2. Hasanpourghadi, M. et al. *Hepatology.* 2020;72(Suppl 1):505A. Presented at the AASLD Meeting 2020 (Abstract 826).

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Methods

Combination HBV PolN, PolC & Core Studies

Vectors investigated*

AdC(6/7)-gDPolN, -gDPolC, -gDCore

- N- or C-terminal part of polymerase or core within gD

AdC(6/7)-gDHBV2 (VRON-0200)

- Polymerase + core within gD

AdC(6/7)-HBV2

- Polymerase + core without gD

AdC(6/7)-gDHBVsd^

- Subdominant regions of N-terminal part of polymerase + C-terminal part of polymerase + core within gD

Analyses of T cell responses

Post-vaccination analyses of T cell responses in blood, spleen, and liver

- Intracellular cytokine staining (ICS) for IFNγ
- MHC I tetramer staining combined with phenotypic analyses
- Epitope mapping of HBV-specific $CD8^+$ T cells by ICS

Challenge experiment

AAV8-1.3HBV AAV vector model – 1 x 10^9 vg IV

Vaccine vectors – Single IM dose of 1 x 10^{10} vp

- Administered 4 weeks after AAV8-1.3HBV injection

*AdC6 and AdC7 are heterologous chimpanzee adenoviral viral vectors of serotype 6 and 7.
^gDHBVsd also referred to as AdC6-gDHBV3 and AdC7-gDHBV3 when combined with AdC6 and AdC7 vectors, respectively.
ICS, intracellular cytokine staining; IV, intravenous; sd, subdominant; vg, viral genome; vp, virus particles.

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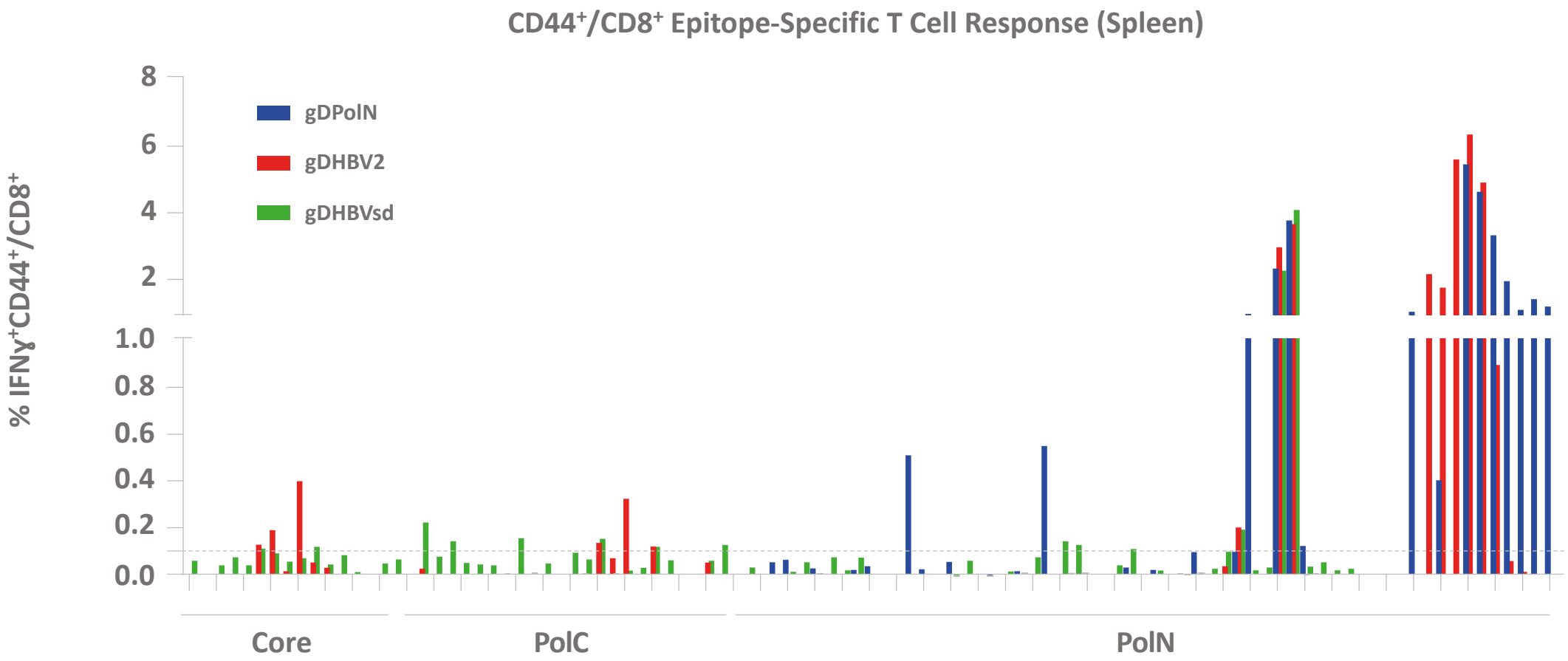
VRON-0200 Induces CD8+ T Cell Responses to Core and Polymerase



Five male C57Bl/6 mice received either a high- or low-dose prime and boost of heterologous AdC. ICS was performed on spleens of sacrificed animals at study completion. Results shown here are from animals administered low dose (high dose shows similar pattern). gDHBVsd also referred to as AdC6-gDHBV3 and AdC7-gDHBV3 when combined with AdC6 and AdC7 vectors, respectively.

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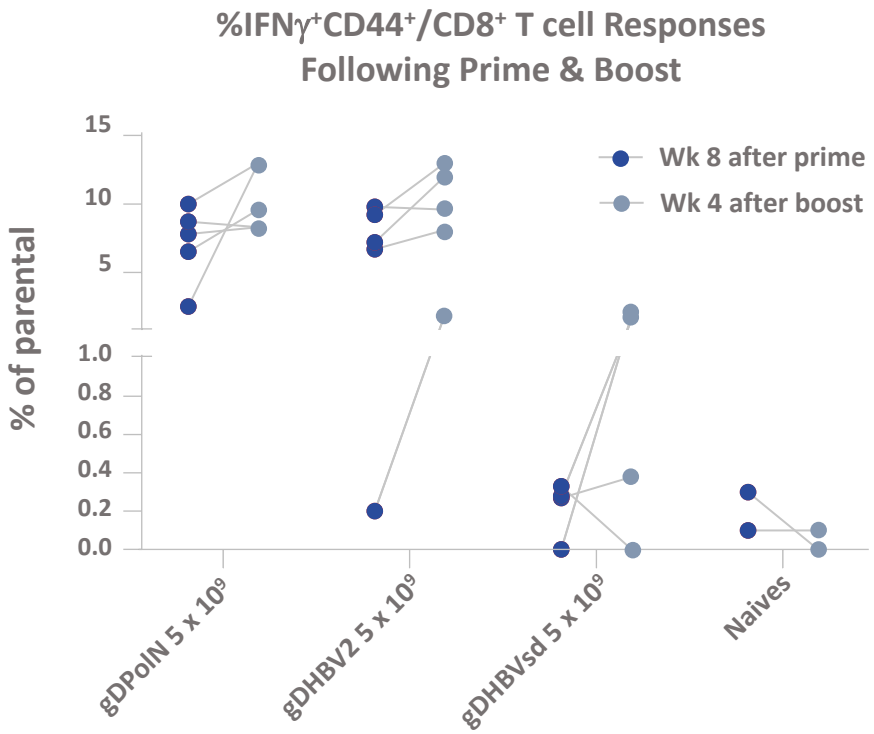
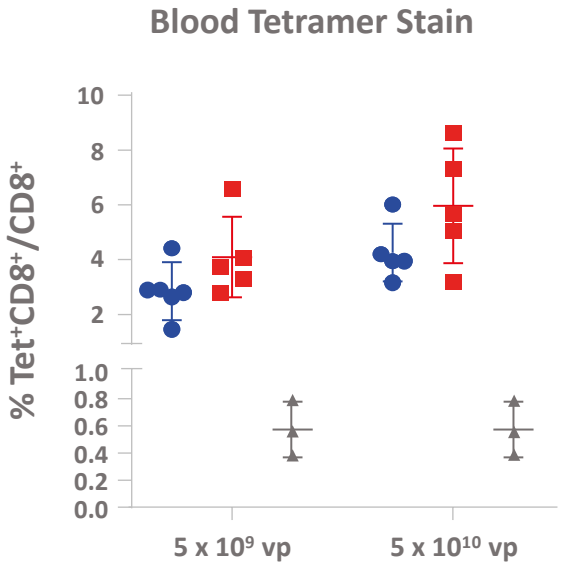
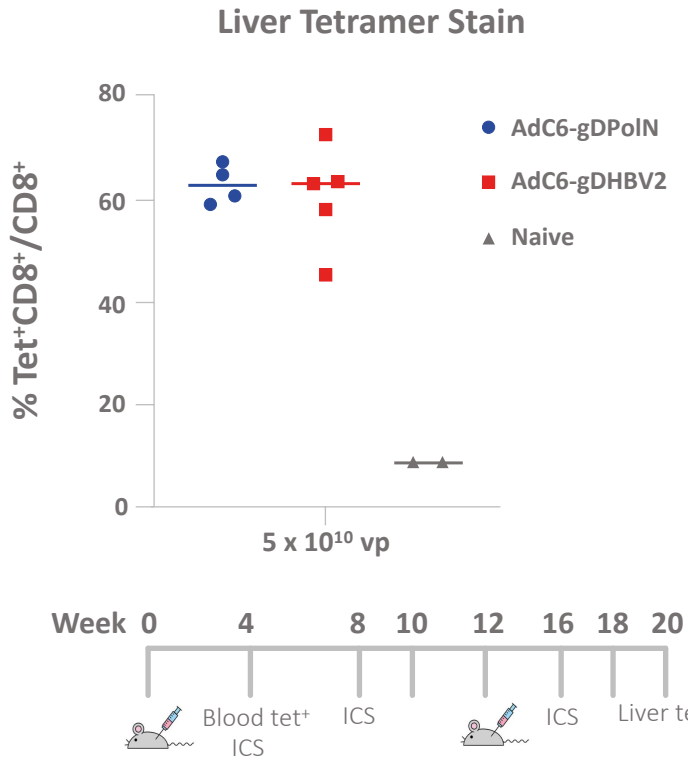
Magnitude of VRON-0200-Induced Immune Responses Similar to Those to gDPolN







T cell responses in individual C57Bl/6 mice were assessed from blood sampled at week 8 after the prime (prior to the boost) and then 4 weeks after the heterologous boost.

CD8+ T cell responses to the dominant polymerase epitope were assessed in C57Bl/6 mice upon staining with an HBV-Pol-specific MHC class I tetramer. Tetramer-binding T cells were detected in blood 4 weeks after a prime and in livers after prime and boost.

tet+, tetramer staining; gDHBVsd also referred to as AdC6-gDHBV3 and AdC7-gDHBV3 when combined with AdC6 and AdC7 vectors, respectively.

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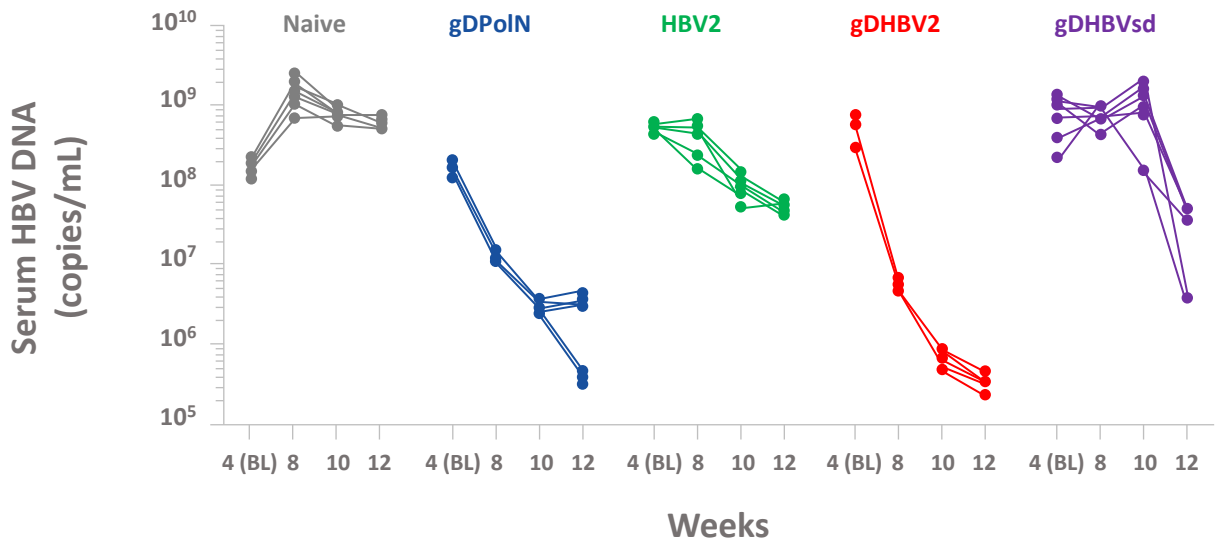
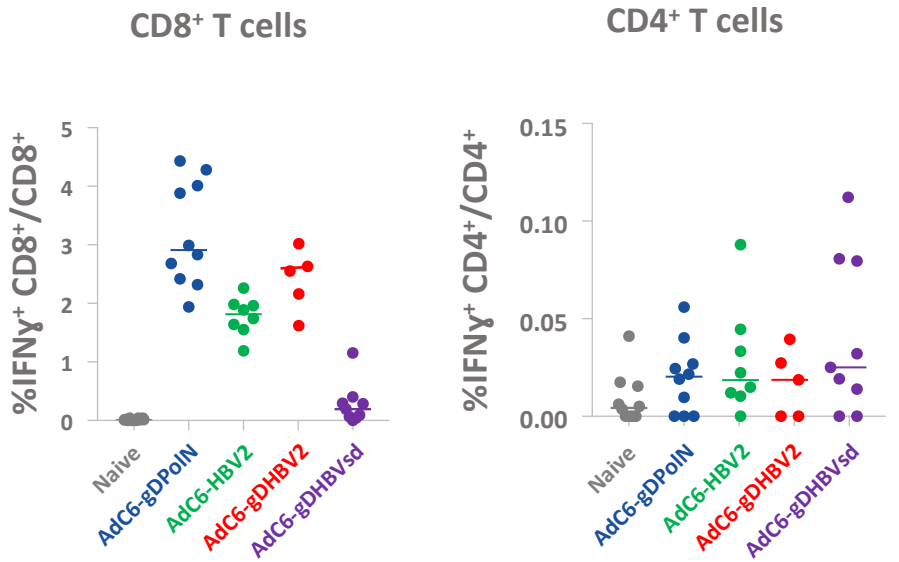
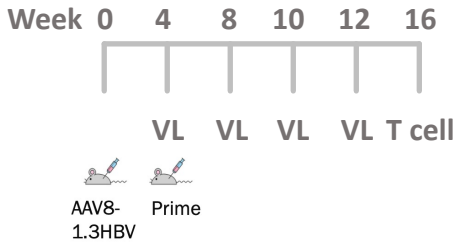
gD Enhances Antiviral Activity, Which Correlates with CD8⁺ T Cell Responses







AAV8-1.3HBV – 1 x 10^9 vg IV at week 0

All vaccines given by IM injection at a dose of 1 x 10^{10} vp at week 4 (n=10 per group); C57Bl/6 mice

Correlations*

r-values	CD8	CD4	vgc
CD8		-0.020	-0.491
CD4	-0.020		0.157
vgc	-0.491	0.157	

P-values	CD8	CD4	vgc
CD8		0.914	0.004
CD4	0.914		0.389
vgc	0.004	0.389	

Spearman Rank Correlation

*Correlations for CD44⁺CD8⁺ T cells: r-value –0.77, *P*<.0001.
BL, baseline; vgc, viral genome copies; VL – viral load.
gDHBVsd also referred to as AdC6-gDHBV3 and AdC7-gDHBV3 when combined with AdC6 and AdC7 vectors, respectively.

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Conclusions

- **VRON-0200 vaccination**
  - Vaccination elicits potent and broad $CD8^+$ T cell responses to HBV core & polymerase
  - Vaccine-induced $CD8^+$ T cells traffic to the liver
  - Multi-log HBV DNA viral load declines after a single IM injection
    - ✓ gD required for optimal antiviral activity
    - ✓ Vaccine-induced $CD8^+$, but not $CD4^+$ T cell responses correlate with antiviral activity
- **A Phase 1b clinical study is planned (Q4 2022)**
  - Study population: HBeAg negative, on nucleos(t)ide therapy
  - Prime only and prime & boost regimens

HBeAg, hepatitis B e-antigen.

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