What's on the Horizon for Treatment for CHB? New Targets for HBV?

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www.vidrl.org.au/publications/hep_updates.htm

Guidelines HBV Treatment

	AASLD 2009	EASL 2012	APASL 2012
Lamivudine	Not preferred	Not preferred	Not preferred
Adefovir	Not preferred	Not preferred	Not preferred
Entecavir	**First line**	**First line**	**First line**
Telbivudine	Not preferred	Not preferred	Not preferred
Tenofovir	**First line**	**First line**	**First line**
PEG-IFN	**First line**	**First line**	**First line**

In many regions the cost of anti-CHB therapy poses significant financial burden to patients and to the resource-constrained national healthcare systems

Financial burden of treatment remains unaffordable for most patients because of lack of full or adequate reimbursement for treatment

Lok and McMahon 2009, Liaw et al., 2012, EASL 2012

Benefits of HBsAg Clearance

- ↓ Hepatic decompensation
- ↓ HCC
- ↑ Survival
- ↓ Levels of cccDNA
- **As close to cure as we can expect to achieve in chronic hepatitis B**

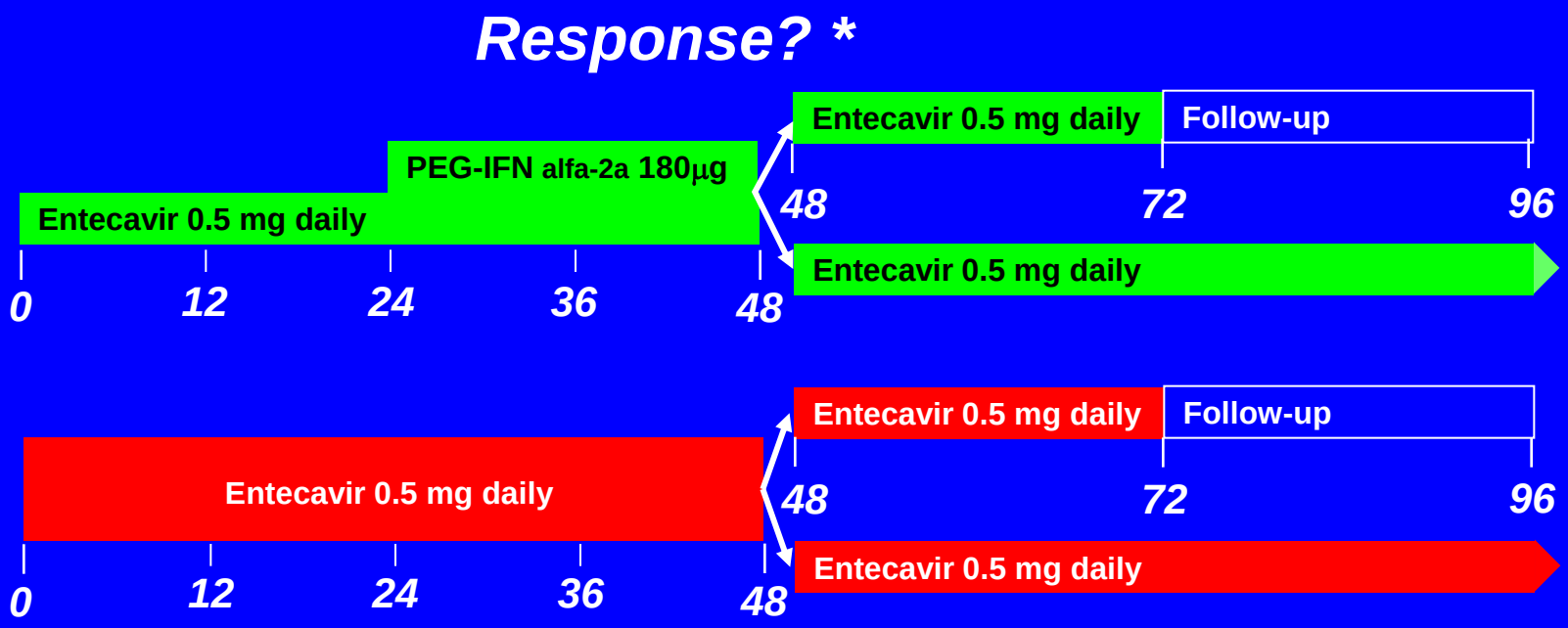
Fattovich G, et al. Am J Gastro 1998; 93:896-900. Werle-Lapostolle B, et al. Gastroenterology 2004; 126(7):1750-1758. Perrillo R. Hepatology 2009; 49:1063-1065.

What's New?

- **Refining current treatments**
  - Combination therapy
    - PEG-IFN + NA
- **New treatment approaches**
  - NA - TAF
  - Novel antivirals
  - Novel immune stimulants

PEG-IFN Add-On (ARES Study)

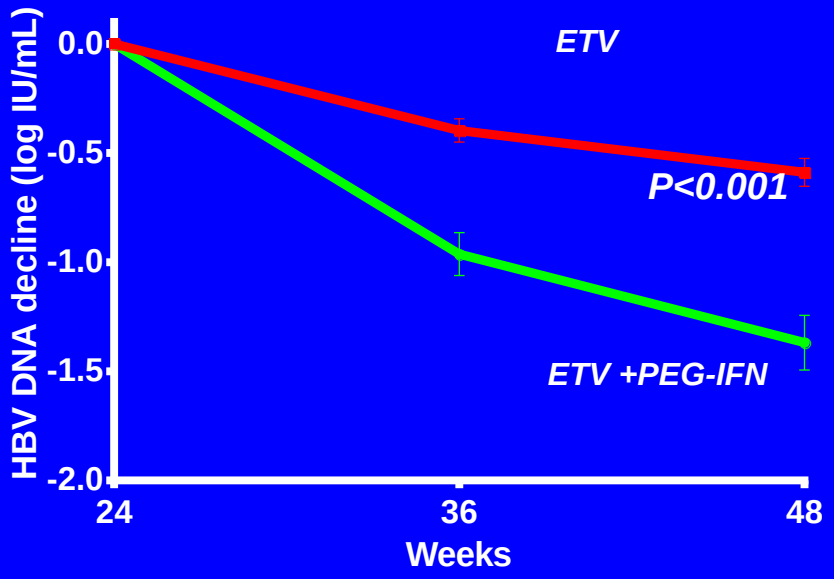
- HBeAg positive study
- Multicenter, open-label, randomized controlled trial

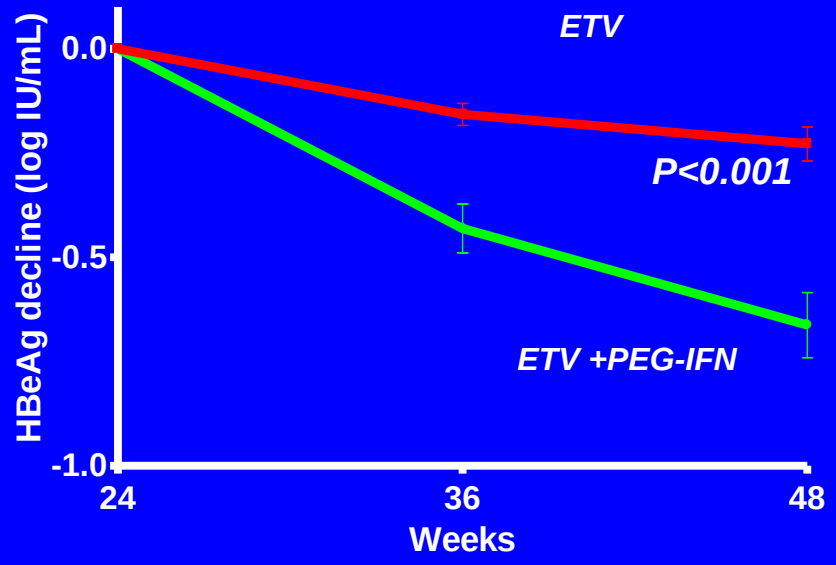


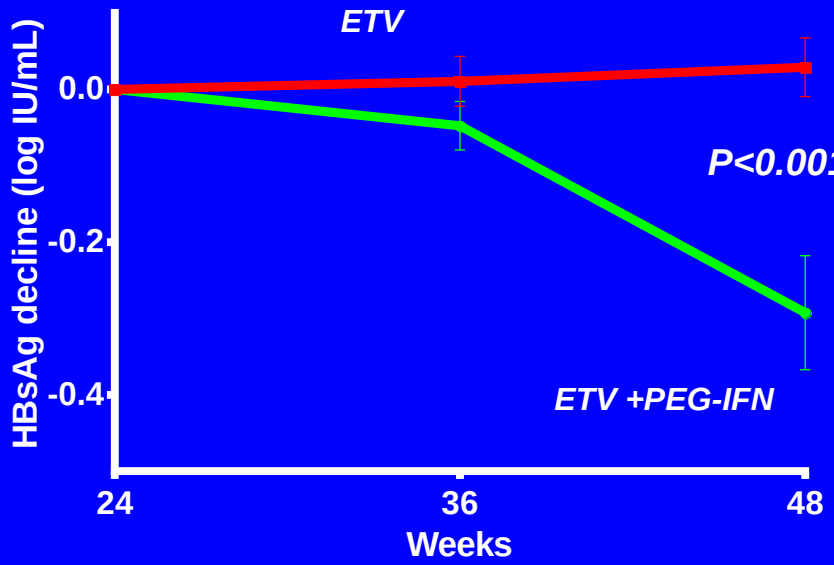
Response: combined presence of HBeAg loss and HBV DNA level < 200 IU/ml at week 48

Sonneveld et al. AASLD 2012

ETV and PEG-IFN (ARES Study): HBV DNA, HBeAg, HBsAg During Therapy



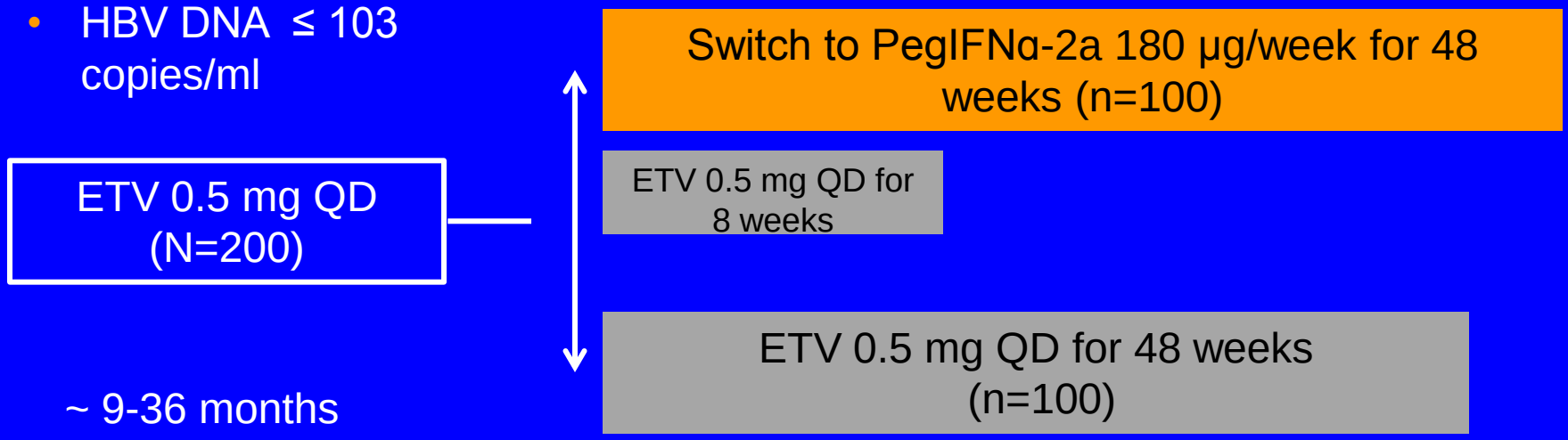




Sonneveld et al. AASLD 2012

PEG-IFN Switch (OSST Study)

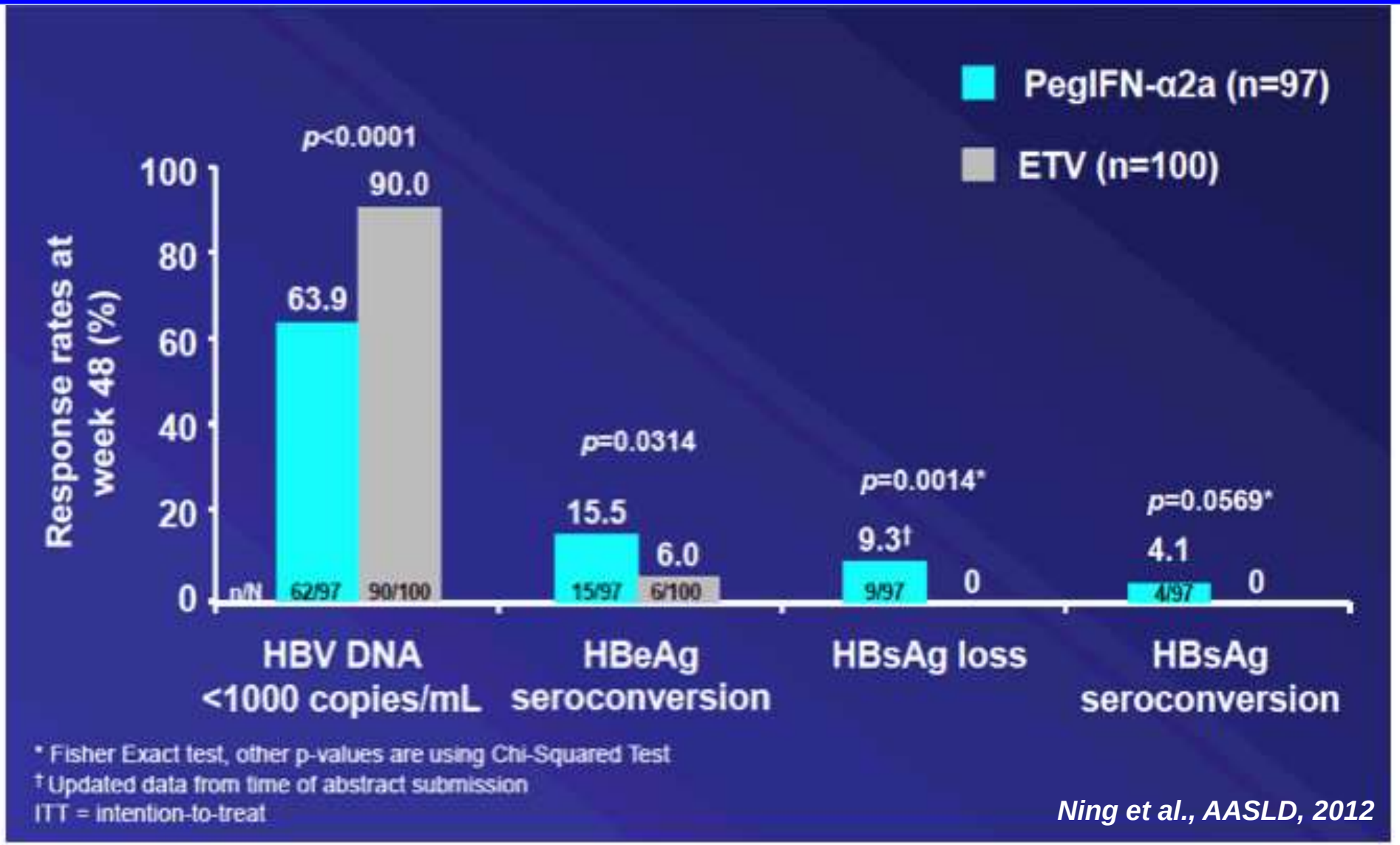
- Randomized, multicentre, open-label study
- Primary endpoint: HBeAg seroconversion at end of treatment (week 48)
- Secondary endpoint: HBsAg loss at week 48



QD = once daily; PEIU = validated with in-house reference standards obtained from Paul Ehrlich

Ning et al., AASLD, 2012

OSST Study: Response Rates at Week 48 of Treatment (ITT)



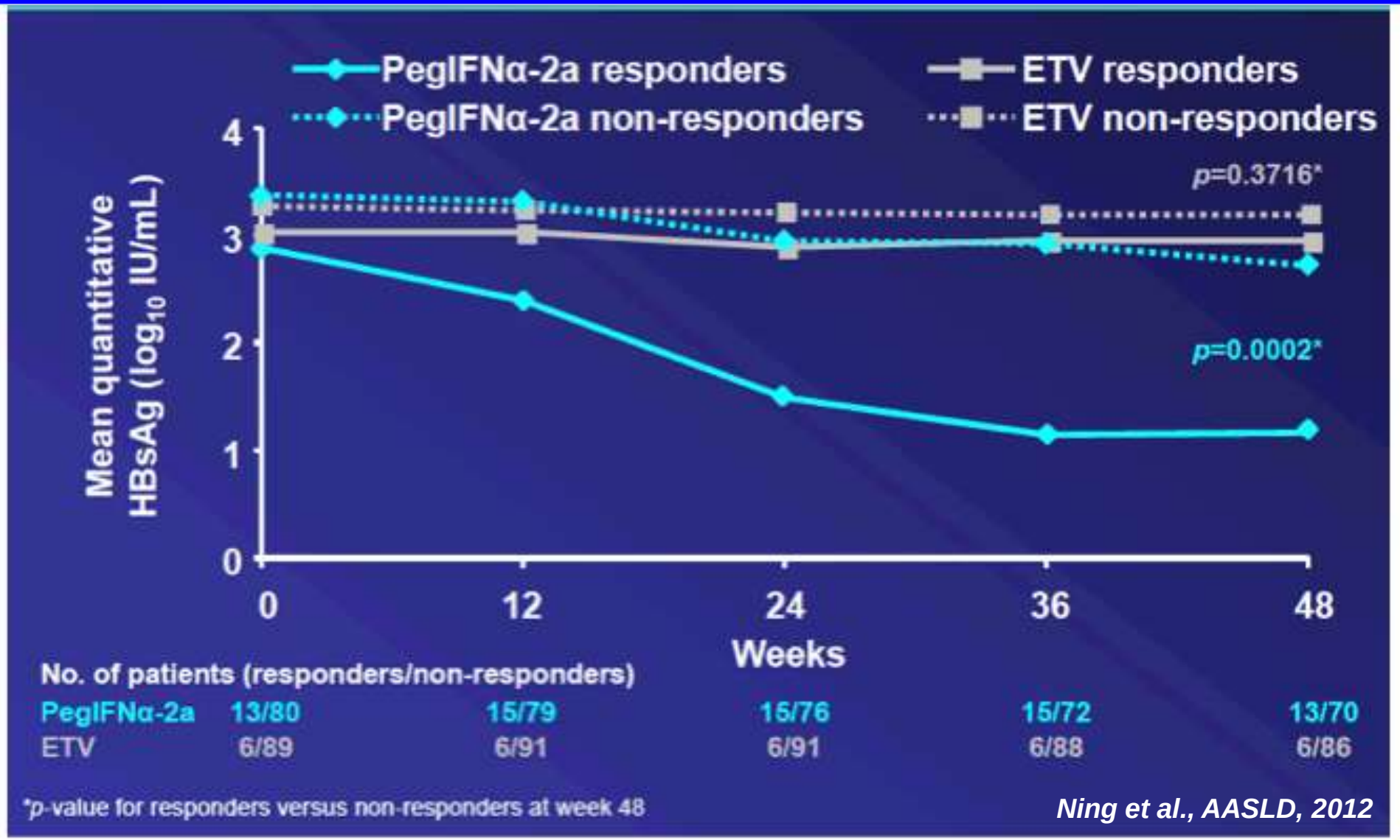
* Fisher Exact test, other p-values are using Chi-Squared Test

† Updated data from time of abstract submission

ITT = intention-to-treat

Ning et al., AASLD, 2012

OSST Study: HBsAg Decline Greater in PEG-IFN Responders

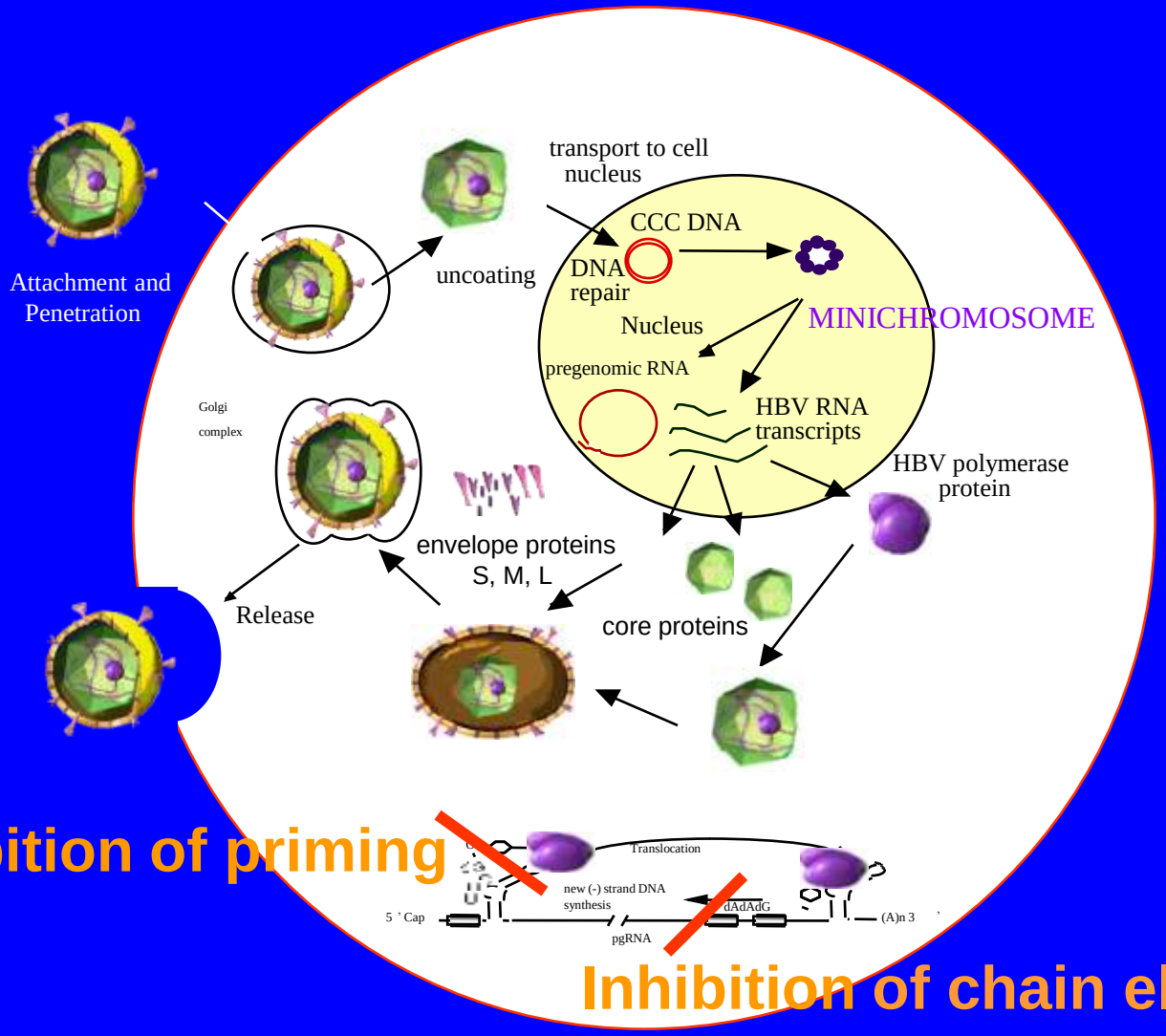


Ning et al., AASLD, 2012

New Treatment Approaches

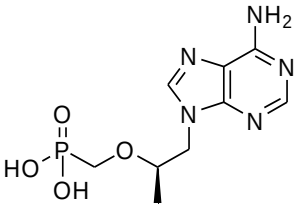
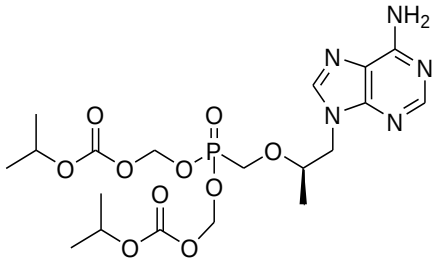
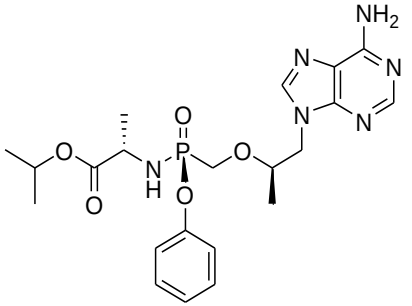
Strategy	Target	Agents		
HBV life cycle	HBV Pol	TAF		
	Viral entry	Myrcludex-B		
	cccDNA	Zinc finger nucleases	cccDNA conversion inhibitors	
	mRNA transcription/ stability	Zinc finger proteins	Epigenetic silencers	RNA silencing -Antisense OGNs -Ribozymes -RNAi
	Viral assembly	HAPs	Phenylpropenamides	
	HBV antigen secretion	REP 9AC’	Small molecule inhibitors of HBsAg secretion e.g. glucovirs e.g. triazolo-pyrimidines	
Immuno-therapeutic	PegIFN-λ1a (IL29)			
	Cytokines	rIL-7	rIL-21	
	TLR agonists	TLR7 (GS-9620)		
	Therapeutic vaccines	Adeno-virus approaches (TG1050)	Tarmogen (GI-13020)	
	Blocking T cell inhibitory receptors	Anti-PD-1 moAB (BMS936558)	Anti-PD-L1 moAb (BMS936559)	
	Intrahepatic blocking of suppressive cytokines / regulatory T cells	TGF-β inhibitors	T reg depletion (e.g. α-CD25, daclizumab)	

Inhibition of HBV Nucleos(t)ide Analogues



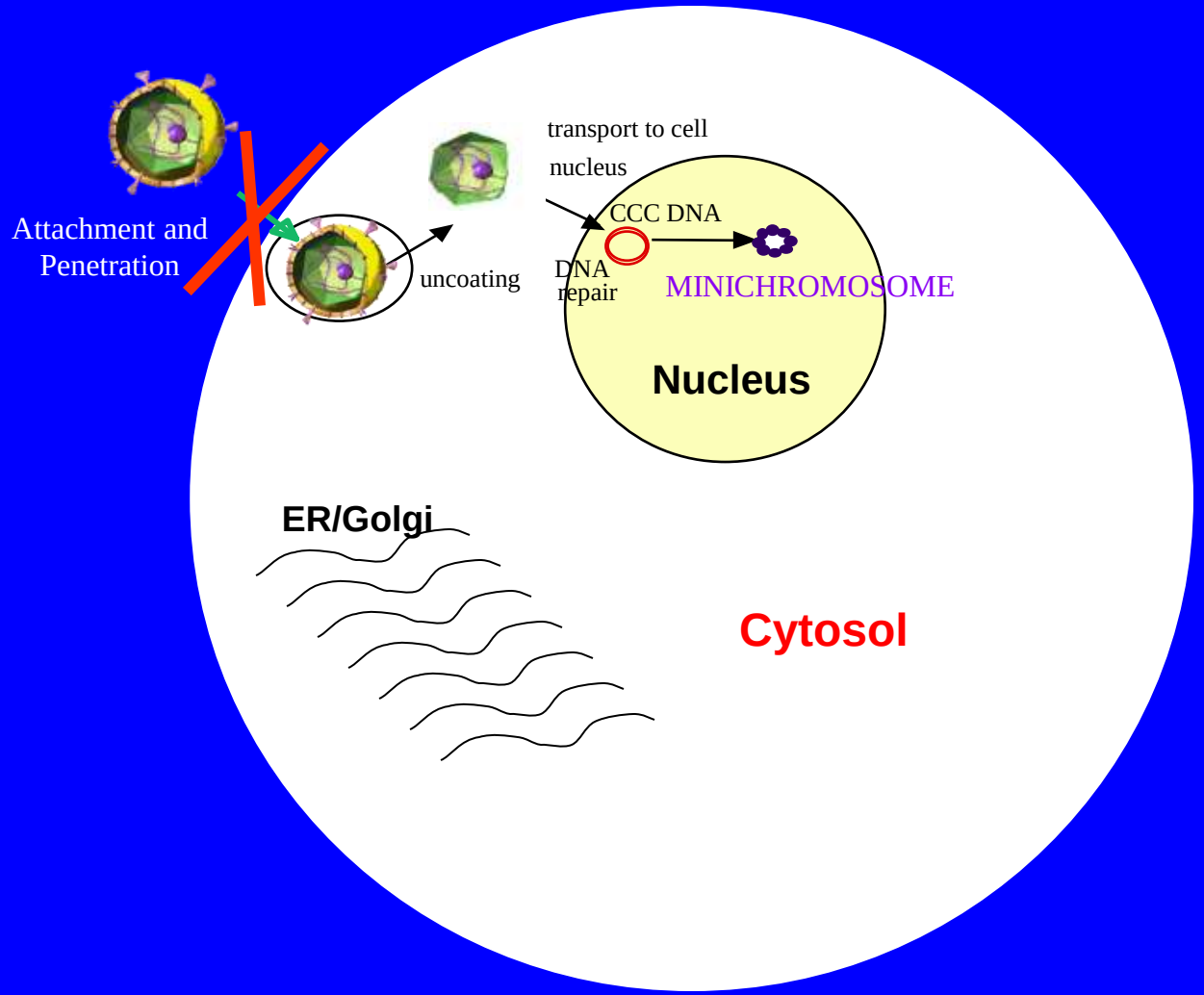
Tenofovir Alafenamide (TAF)

- TAF = orally bioavailable phoshonoamidate prodrug of tenofovir (TDF)
- In comparison with tenofovir, TAF enables enhanced delivery of the parent nucleotide and its active diphosphate metabolite into lymphoid cells and hepatocytes.
- This is attributed to an improved plasma stability and differential intracellular activation mechanism for TAF relative to TDF

EC_{50} HIV-1 (PBMCs)	Tenofovir	Tenofovir Disoproxil	TAF
			
	1.2 µM	0.015 µM	0.003 µM

Prevention of Infection: Entry Inhibitors

Acylated Pre-S1 Peptides



Petersen J, Urban S. et al 2008. Nature (Biotechnology);26:335

eLIFE
elife.elifesciences.org

Sodium taurocholate cotransporting polypeptide is a functional receptor for human hepatitis B and D virus

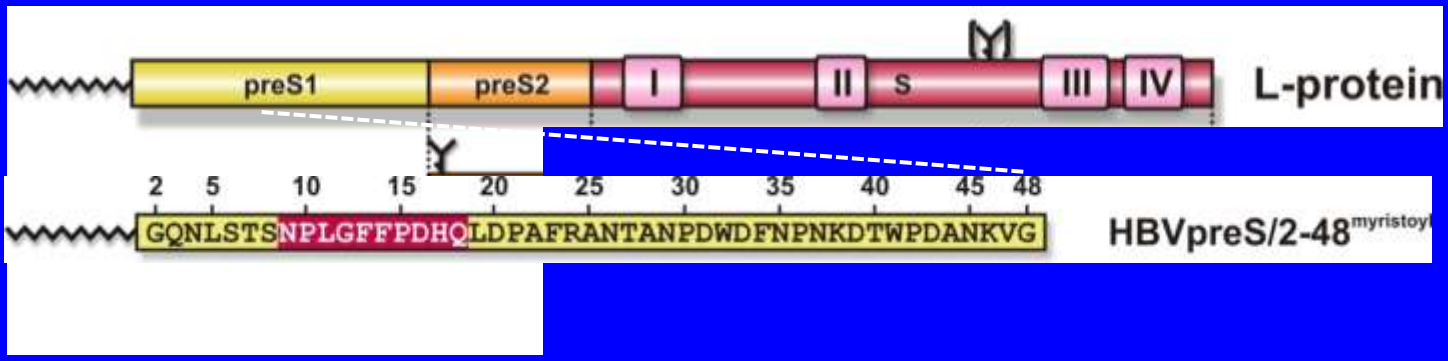
Huan Yan[1,2†], Guocai Zhong[2†], Guangwei Xu[2], Wenhui He[2,3], Zhiyi Jing[2], Zhenchao Gao[1,2], Yi Huang[2,3], Yonghe Qi[2], Bo Peng[2], Haimin Wang[2], Liran Fu[2,3], Mei Song[2,3], Pan Chen[2,3], Wenqing Gao[2], Bijie Ren[2], Yinyan Sun[2], Tao Cai[2], Xiaofeng Feng[2], Jianhua Sui[2], Wenhui Li[2*]

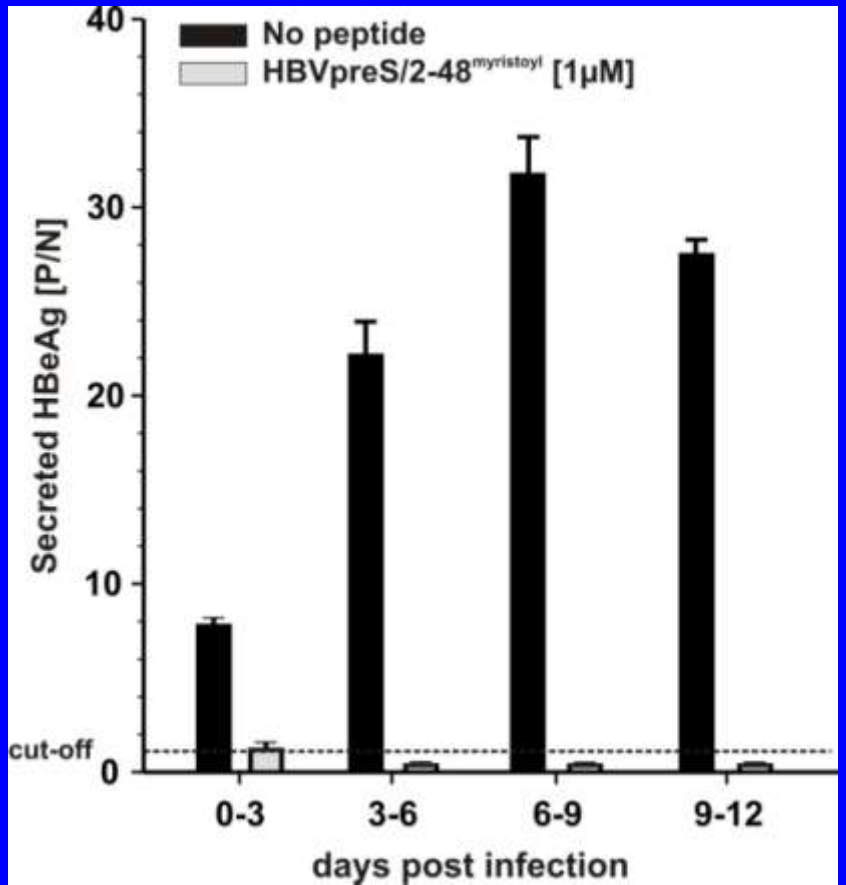
[1]Graduate program in School of Life Sciences, Peking University, Beijing, China; [2]National Institute of Biological Sciences, Beijing, China; [3]Graduate program in Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing, China

NTCP = a receptor for HBV

**Yan et al, eLife, November 2012*

A Synthetic Peptide Derived from the Large Envelope Protein of HBV Blocks HBV Infection in Susceptible Cells….





Infection of HepaRG cells or PHH

Incubation o/n at 37° C

Collection of supernatant of days 8-12p.i.

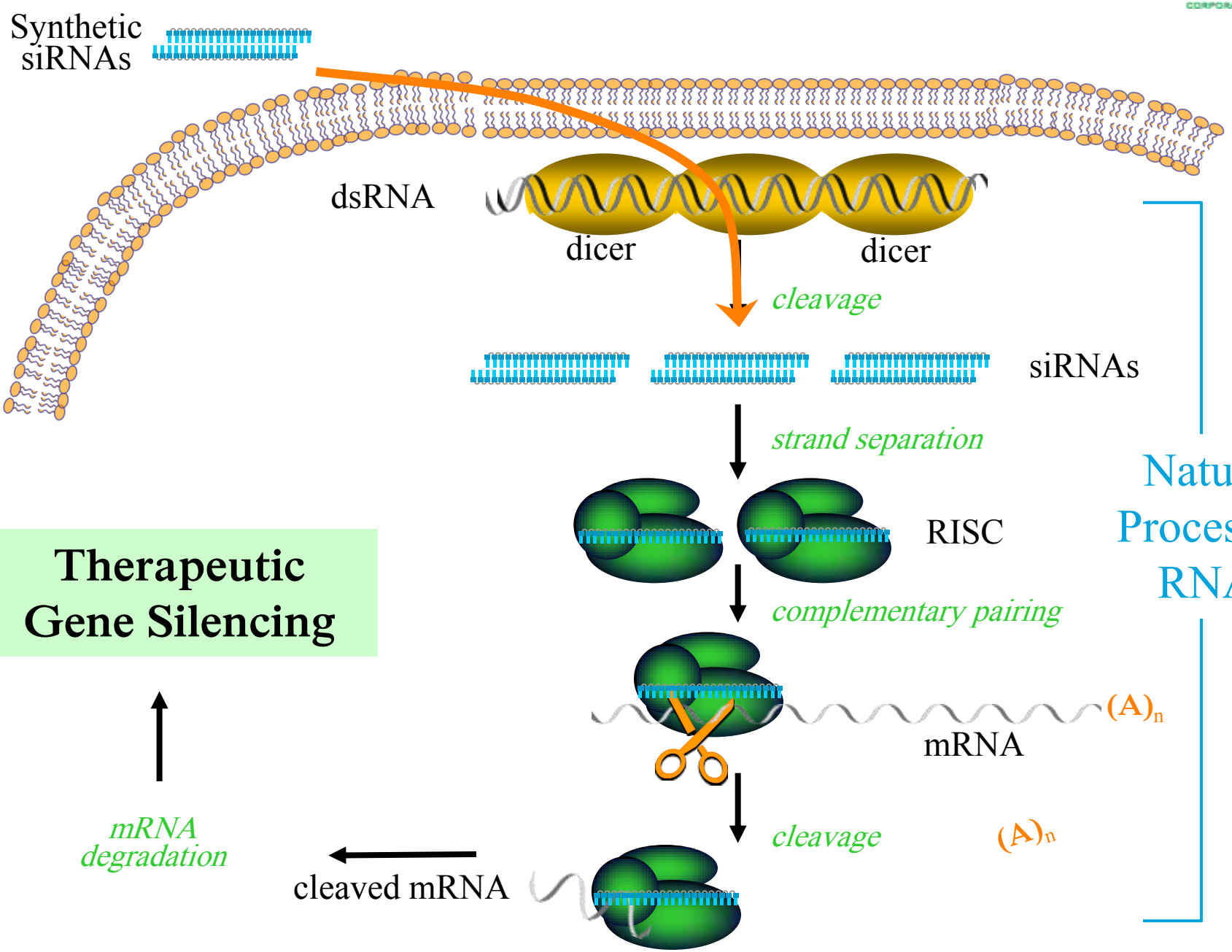
Measurement of secreted HBsAg/HBeAg etc.

Gripon et al., PNAS, 99 (24) 2002
Urban et al., J. Virol, 79 (3), 2005
Glebe et al., Gastroenterology, 129, 2005
Engelke et al., Hepatology, 43, 2006
Schulze et al., Hepatology, 46, 2007

Status of Myrcludex B the First in Class Entry Inhibitor of HBV and Hepatitis Delta Virus (HDV).

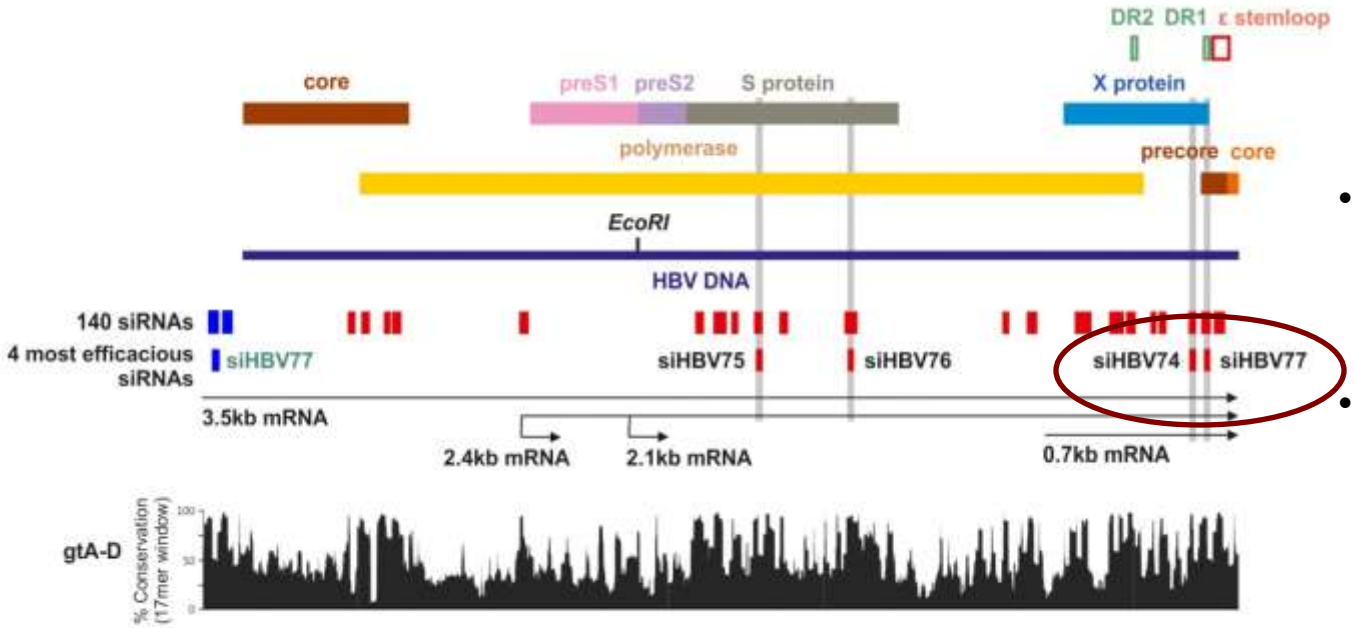
- The GMP synthesis of 100 g Myrcludex B (API) is finished.
- A formulation for s.c. application has been developed.
- Vials for clinical studies have been filled.
- Myrcludex B has been characterized for purity, stability etc.

Mechanism of RNA Interference (RNAi)

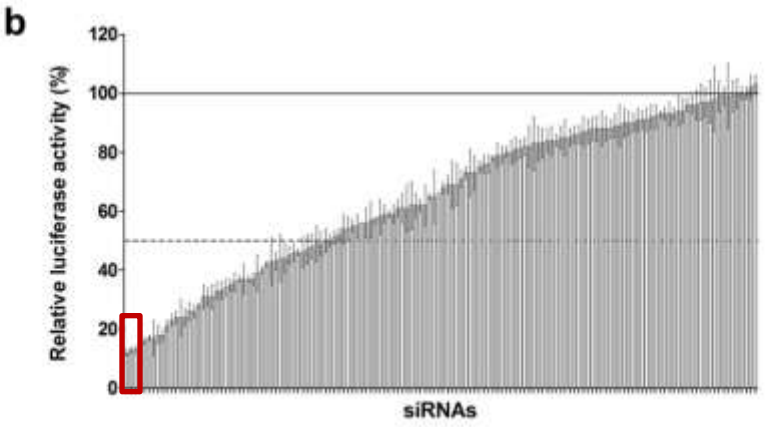


RNAi treatment for chronic Hepatitis B

siRNA design and in vitro screening



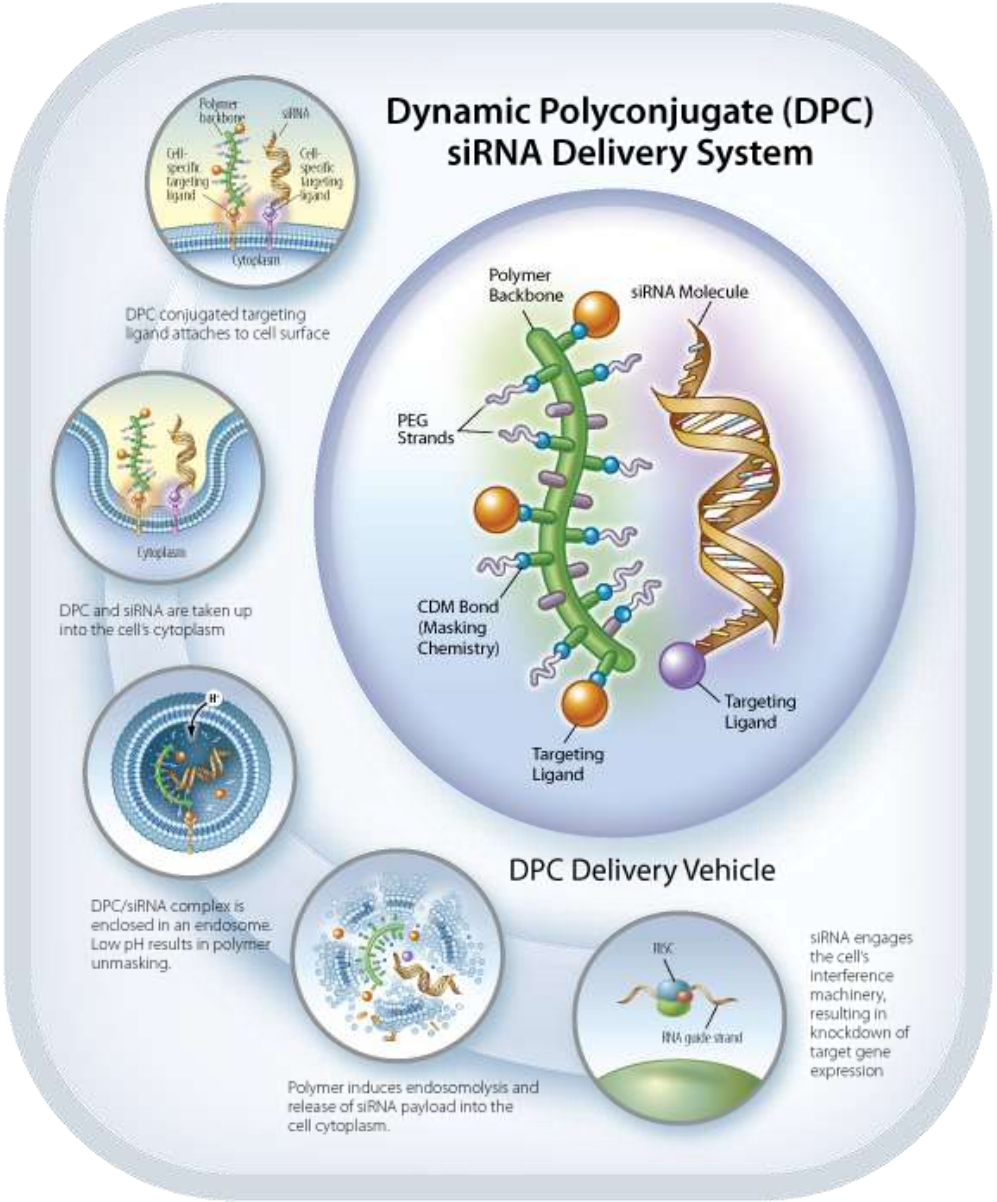
- Designed 140 siRNAs targeting conserved regions of HBV genotypes A-D
- Confirmed conservation in genotypes E-H as well.



- Screened candidate siRNAs in a cell culture system
- 4 highly potent siRNAs chosen for further testing in animal models
- siHBV-74 and siHBV-77 chosen as leads

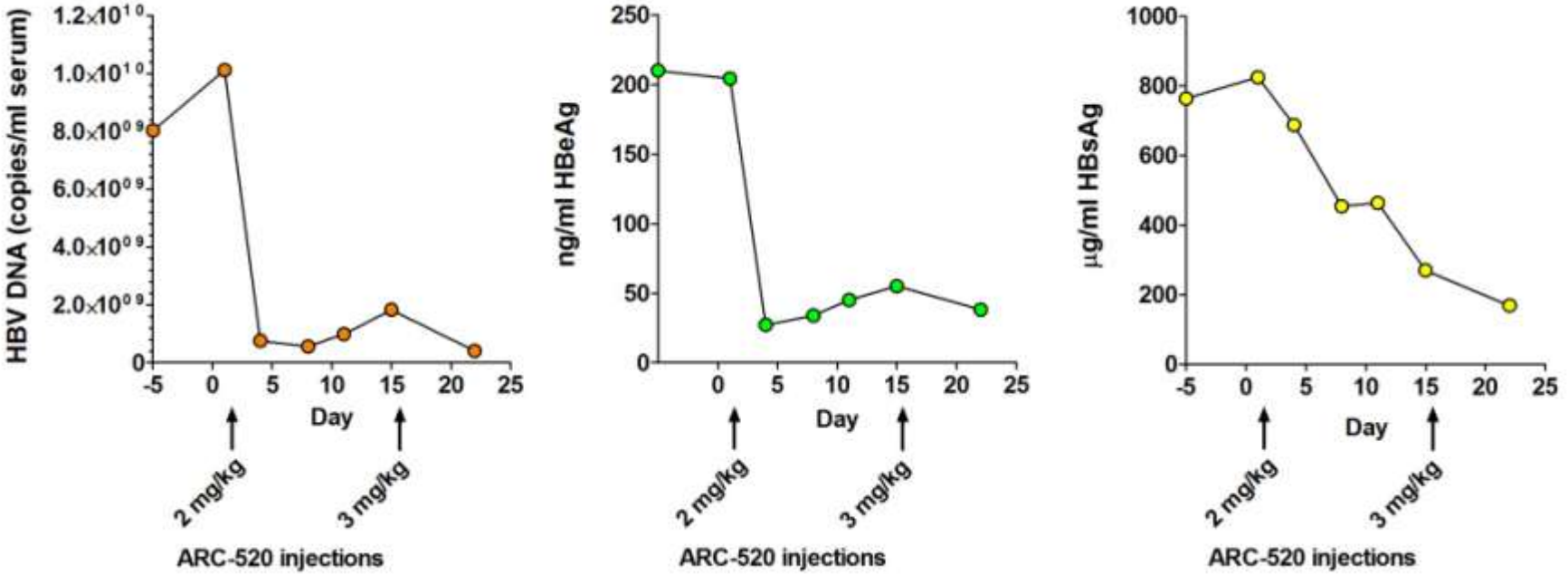
Roche-Kulmbach (Axolabs GmbH)

Dynamic Polyconjugate (DPC) technology for siRNA delivery *in vivo*



- **DPC polymer composition and physical characteristics**
  - Amphipathic peptide
  - peptide amines reversibly "masked" with CDM
  - Slightly negatively charged
- **Cellular uptake of peptide is ligand-driven (N-acetyl galactosamine (NAG)) for hepatocytes)**
- **siRNA is made liver tropic by attachment of lipophilic ligand (e.g. cholesterol)**
- **↓ pH in endosomes drives peptide unmasking**
- **Unmasked peptide disrupts endosomal membrane**
- **siRNA released to cytoplasm**

Reduction in HBV after administration of ARC-520 in a chronically infected chimp



- Log_{10} reduction in HBV DNA (95%), HBeAg (90%) and HBsAg (90%)
- First demonstration of RNAi efficacy in the chimp HBV model
- KD comparable to that achieved in mouse HBV models at similar dose level
- Further reduction after a subsequent dose

REVIVAL OF IMMUNE RESPONSES AND FUNCTIONAL CURE

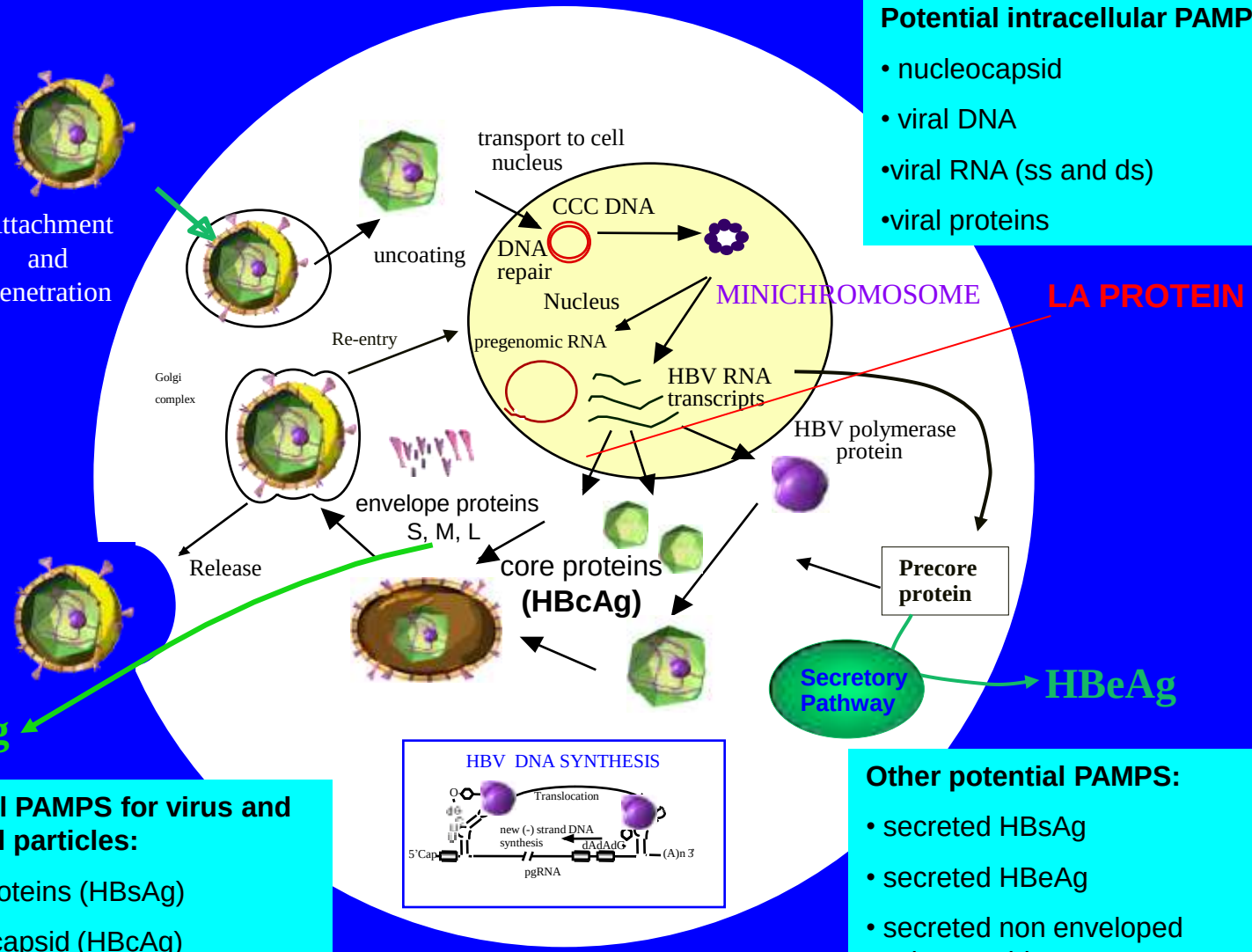
Dr. Robert Lanford, Texas Biomedical Research Institute

Immune-Based Therapies for Chronic Hepatitis B

HBV Life Cycle and Innate Immunity

Potential intracellular PAMPS:

- nucleocapsid
- viral DNA
- viral RNA (ss and ds)
- viral proteins



Potential PAMPS for virus and sub-viral particles:

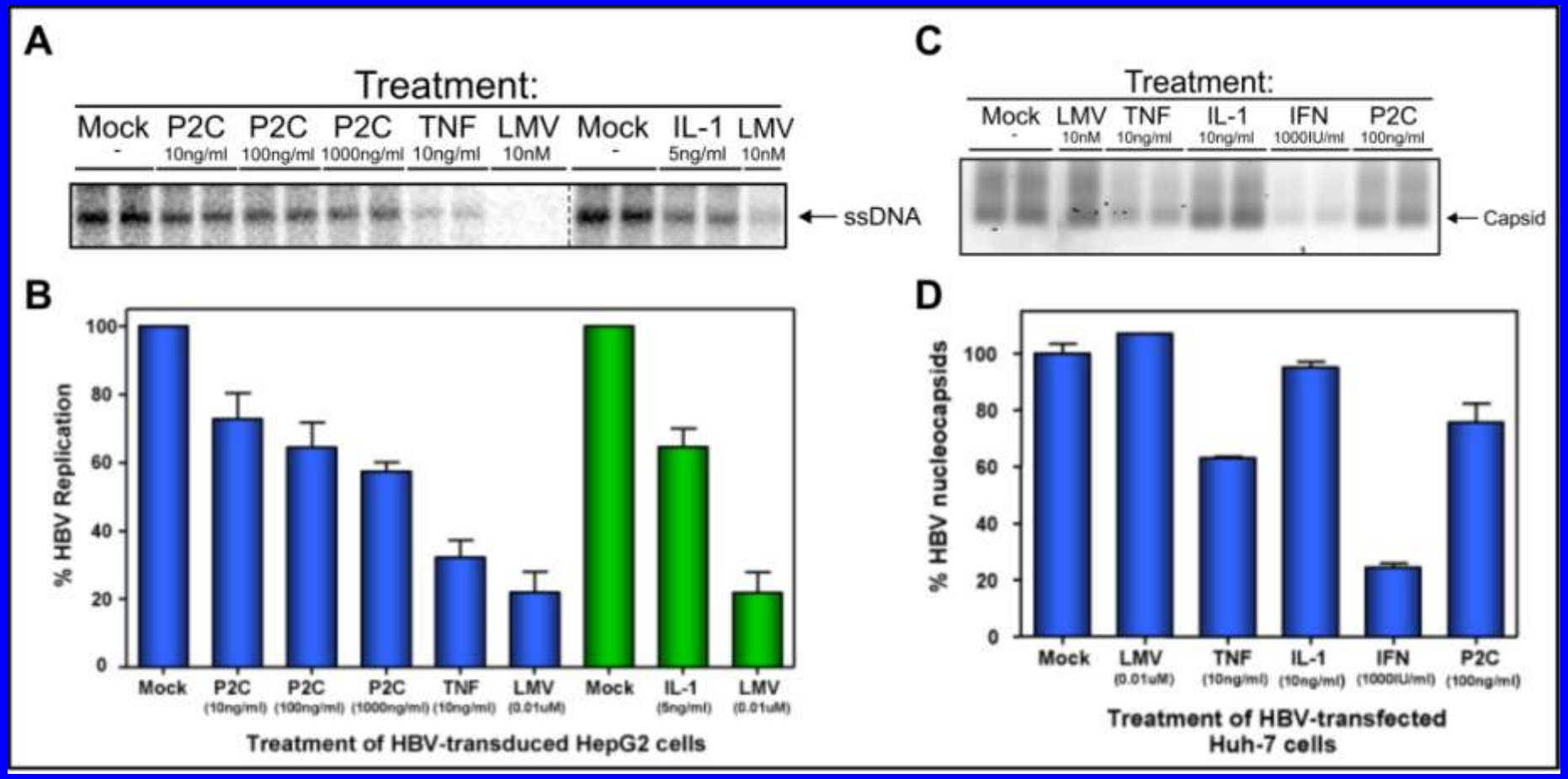
- glycoproteins (HBsAg)
- nucleocapsid (HBcAg)
- rcDNA

Other potential PAMPS:

- secreted HBsAg
- secreted HBeAg
- secreted non enveloped nucleocapsids
- free viral nucleic acids

Modified from Ait-Goughoulte, M. et al 2010. Viruses;2:1394-1410

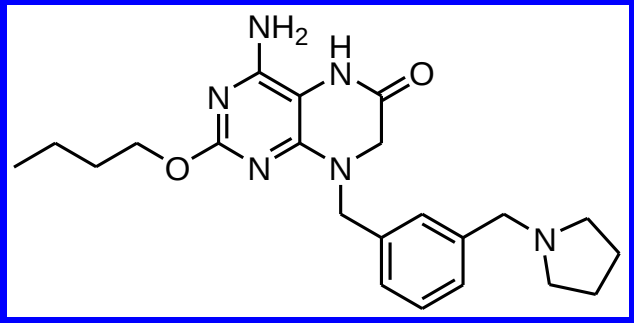
TLR2 Stimulation Inhibits HBV Replication: WT (HBeAg-Positive) HBV (*in vitro*)



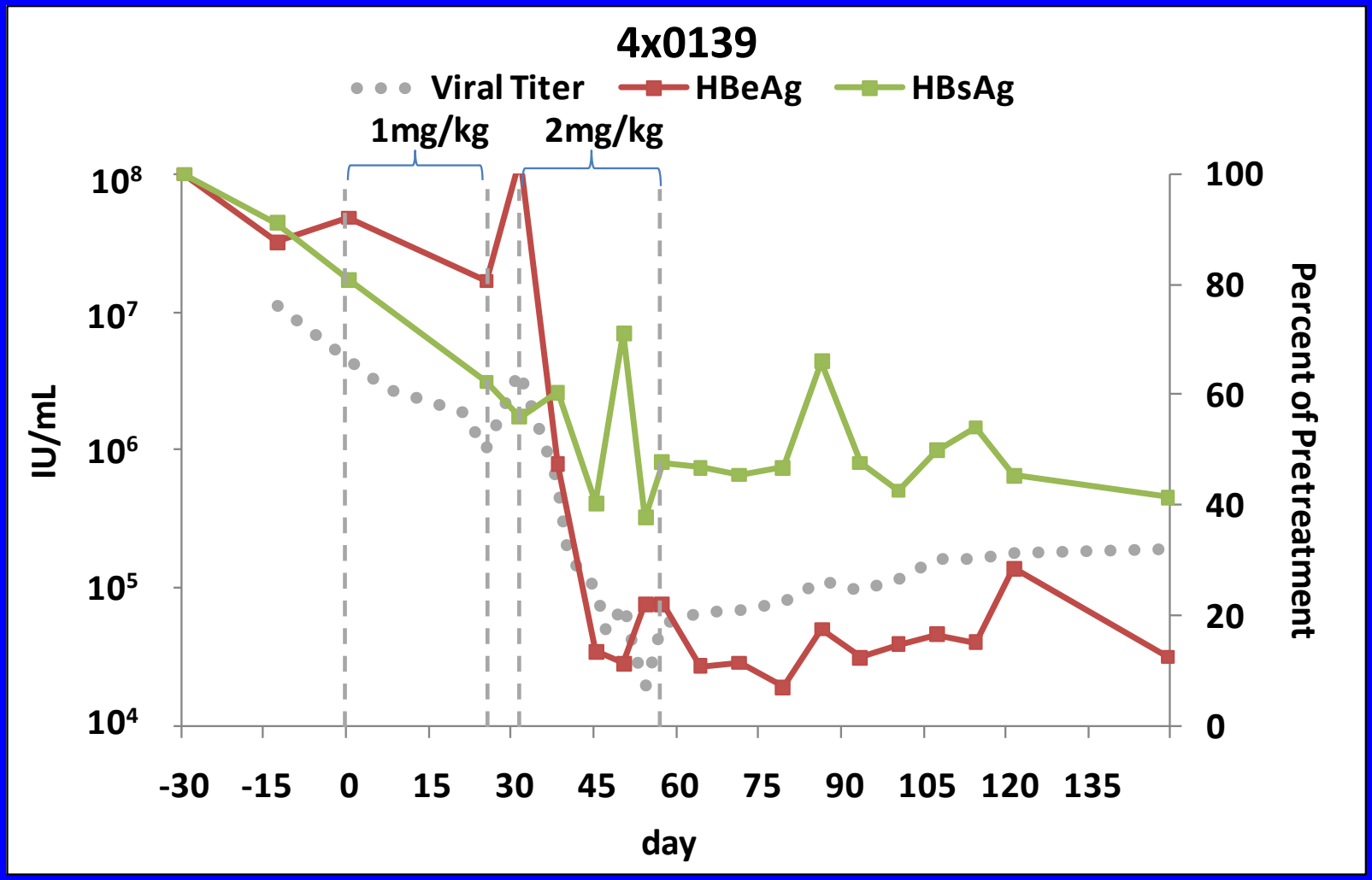
Thompson, A et al 2009. Antiviral Therapy; 14:797–808

GS-9620: Oral TLR-7 Agonist

- TLR-7
  - Intracellular pathogen sensor
    - endolysosomal RNA
  - Agonism induces anti-viral response via innate immune activation
- GS-9620
  - Oral
  - Nanomolar potency
  - Selective (TLR-7 >>> TLR-8)
  - Pharmacodynamic effects in mouse, woodchuck, cyno, chimp, human



GS-9620: Reduction in HBV DNA, and Serum HBsAg and HBeAg in Chimpanzee



Lanford et al. EASL 2011

Reversal of Immune Exhaustion

i. Role of Immune Regulatory Receptors

- in CHB, immune regulatory receptors (IRR) are the key drivers of T-cell dysfunction [eg: PD-1]

  (Fisicaro, P et al 2010. Gasto;138:682-693.,

  Fisicaro, P et al 2012. Gastro; 143(6):1576-1585.e4)

- blocking these inhibitory IRRs has the potential to restore T-cell function [eg: anti-PD-1/PD-L1]

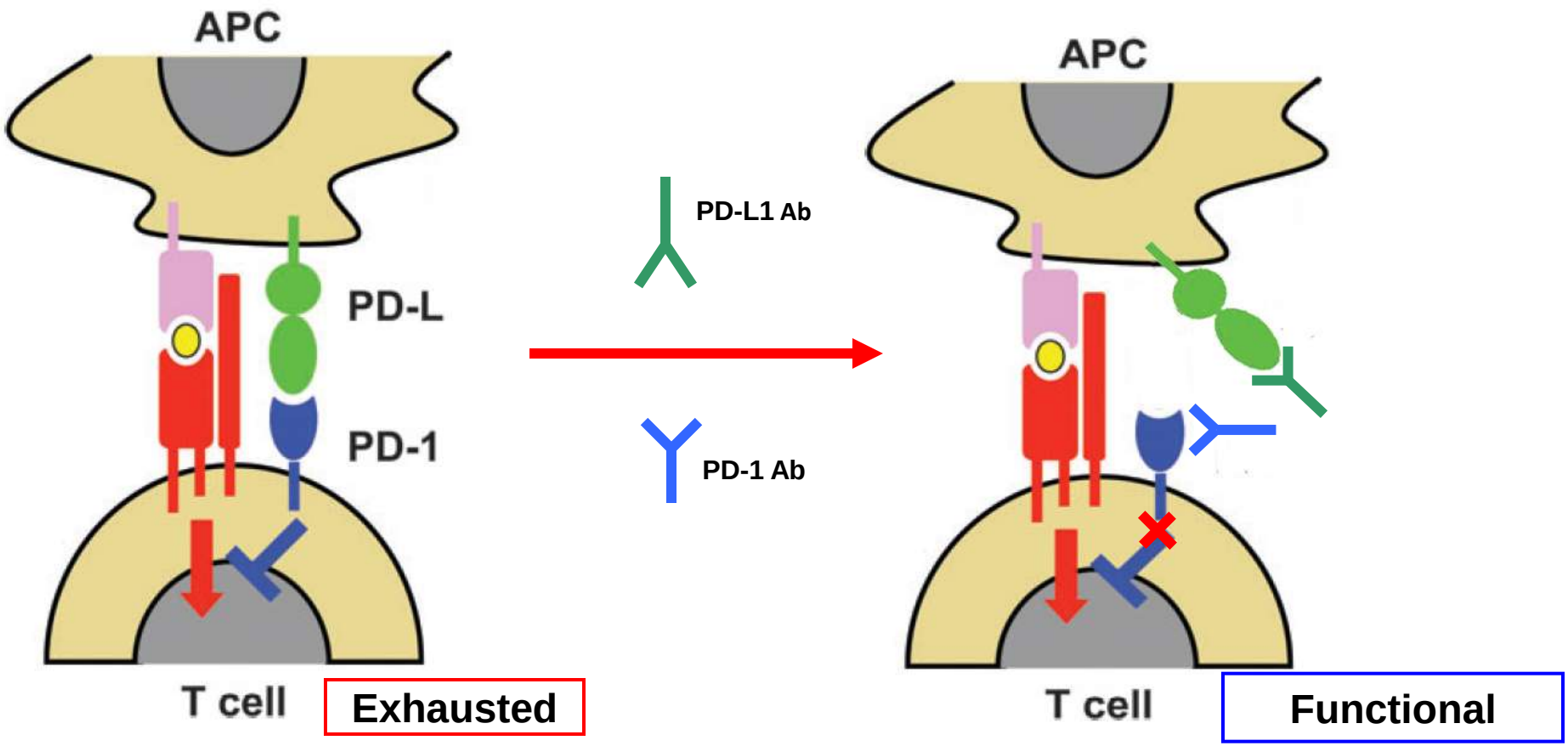
  (Robert, C et al 2013. Euro J Cancer; 49(14):2968-71)

ii. Follicular Helper T-Cells (Tfh)

- Tfh ($CXCR5^+$ $CD4^+$) under influence of IL-21 provide help to B-cells
- IL-21 levels associated with HBeAg seroconversion

  (Ma, S-W et al 2012. J Heapatol;56:775-781)

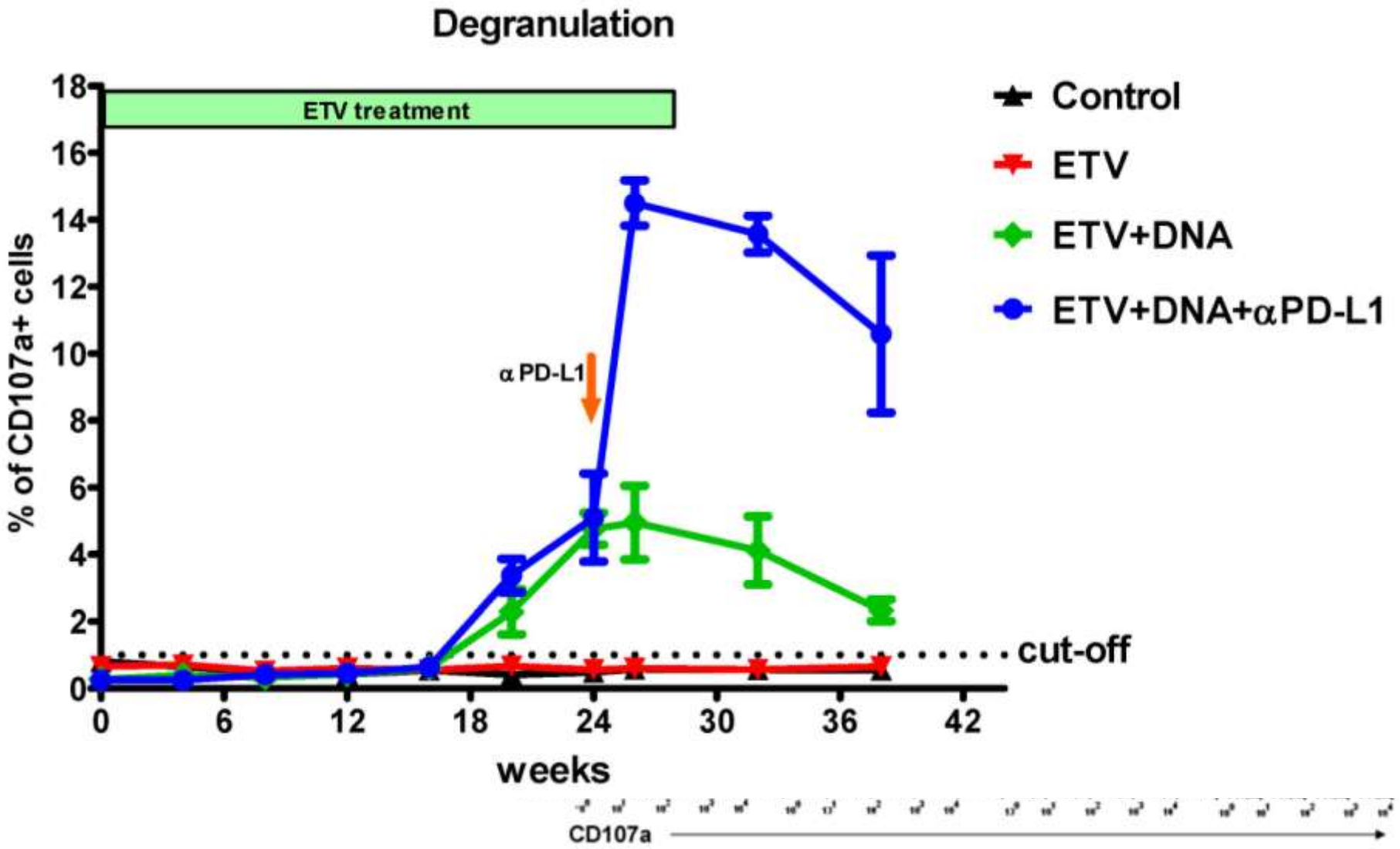
Reverse T cell exhaustion by PD-1/PD-L1 pathway blockade



↓ Proliferation
↓ Cytokine secretion
↓ Cytotoxicity

↑ Proliferation
↑ IFN-γ, TNF-α, IL-2
↑ Cytotoxicity

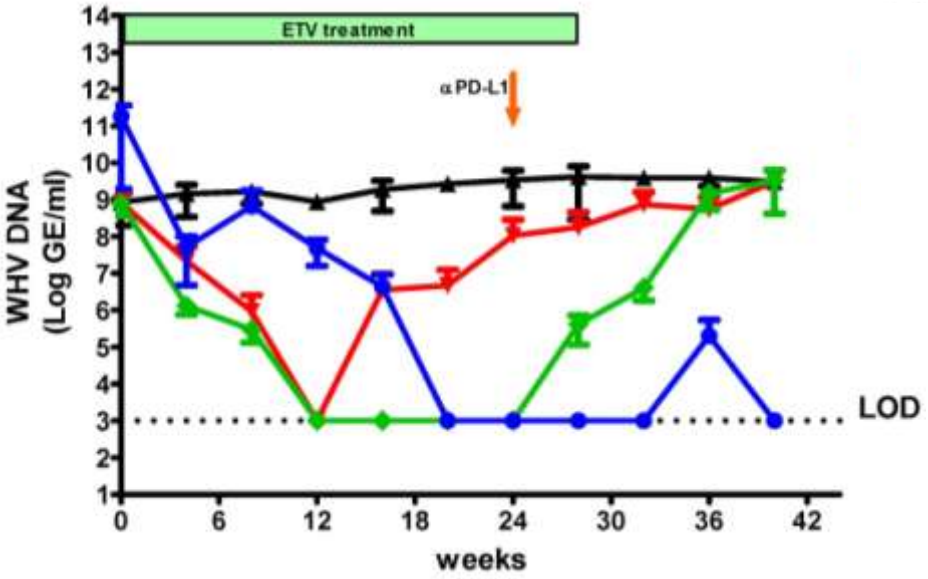
In vivo PD-L1 blockade synergizes with therapeutic vaccination to enhance WHcAg-specific T cell immunity



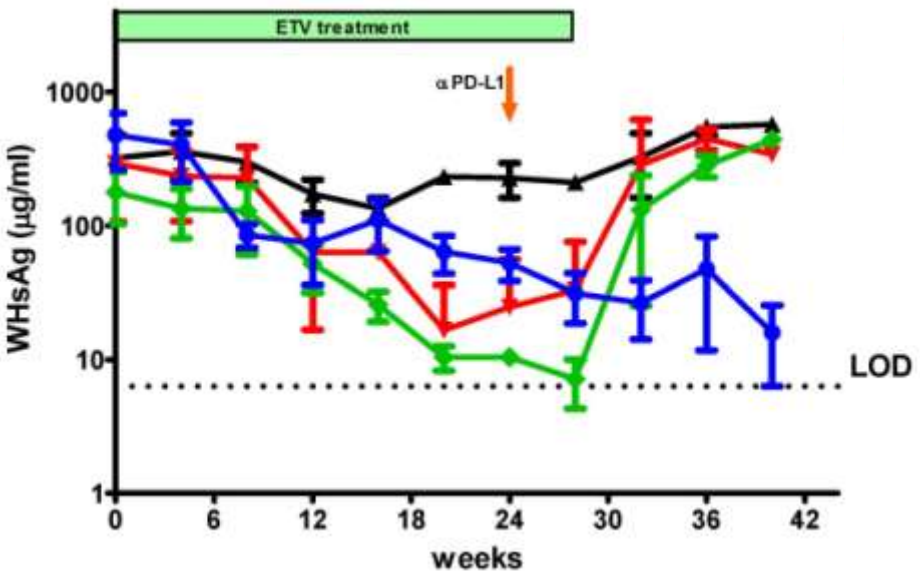
Jia Liu et al, PLOS Pathogens under revision

***In vivo* PD-L1 blockade synergizes with therapeutic vaccination to control WHV replication.**

DNA



WHsAg

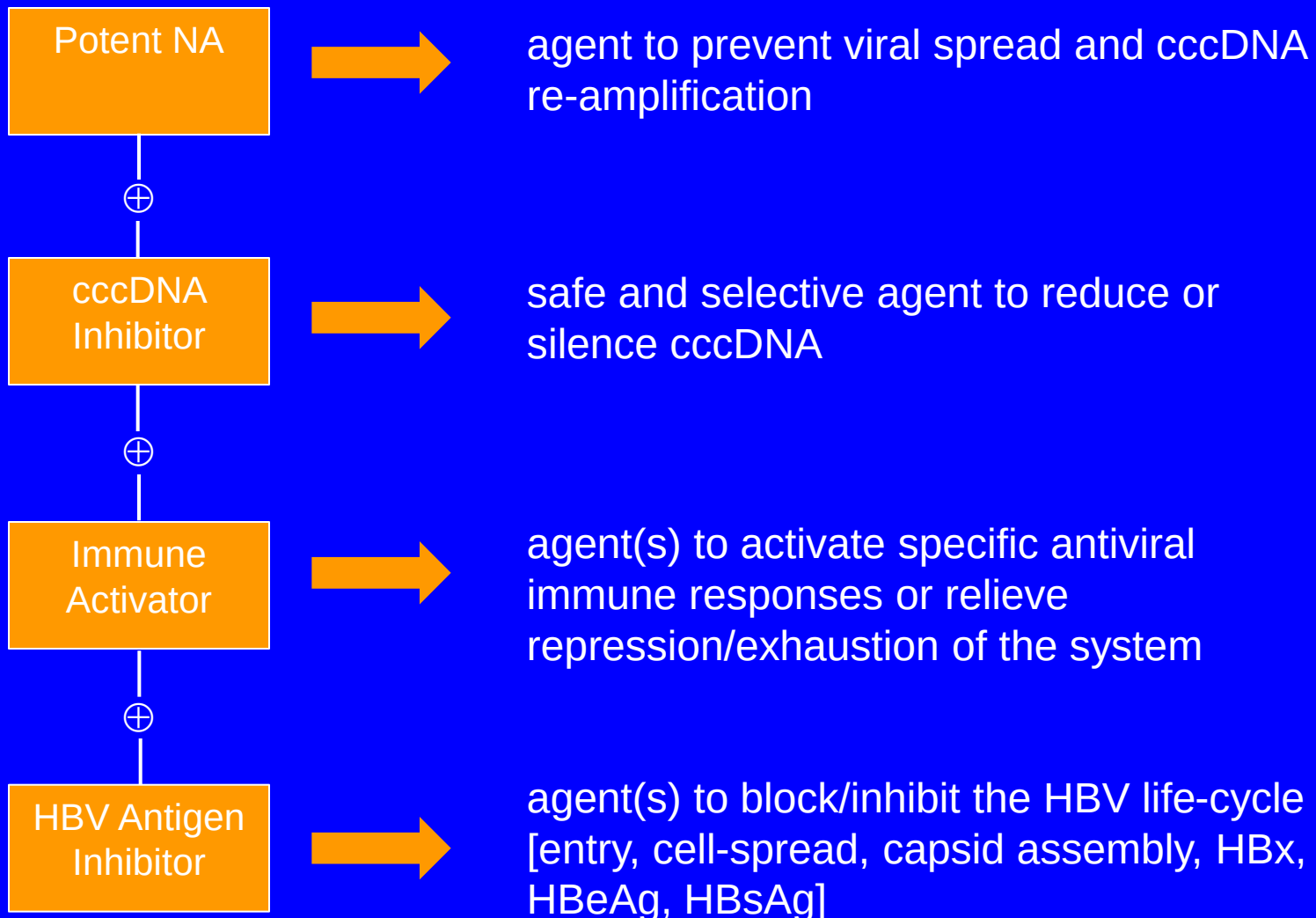


Control
ETV
ETV+DNA
ETV+DNA+αPD-L1

Control
ETV
ETV+DNA
ETV+DNA+αPD-L1

Jia Liu et al, PLOS Pathogens under revision

What Might a HBV Curative Regimen Look Like?



Future Perspectives

- Futility rules for PEG-IFN therapy identified
  - RGT needs to be explored
- The goalposts are shifting
- The long-term aim for the field is to achieve “cure”
  - HBsAg seroconversion
  - An immunomodulator is likely to be required
- Emerging strategies explore the use of combination, or “add-on” PEG-IFN plus NA therapy
- New agents for CHB are starting to emerge
  - Identification of the HBV-R (NTCP) may be paradigm shifting
  - Improved delivery to the liver for molecular therapeutics

PALPABLE OPTIMISM