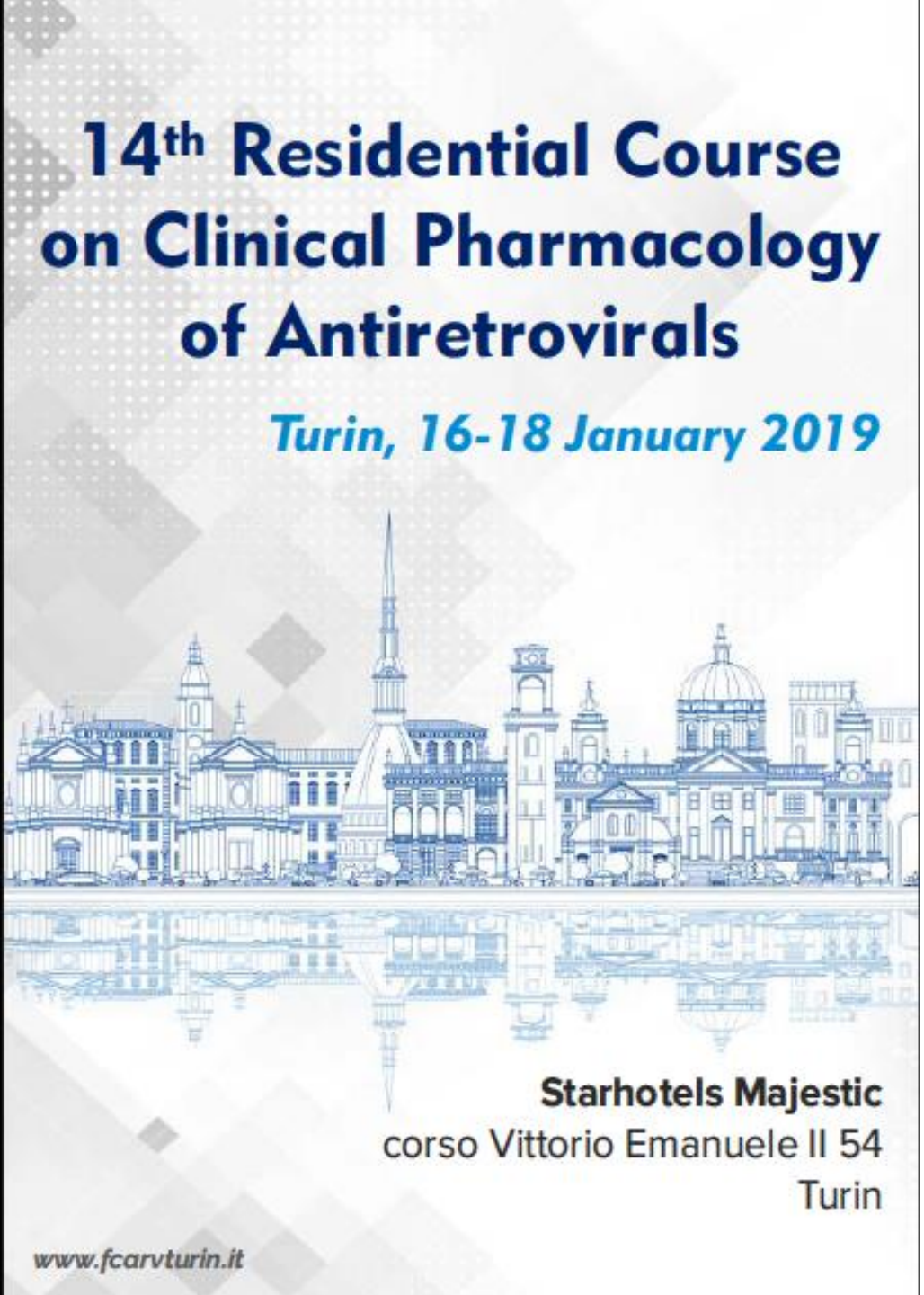

Pipeline and new strategies for HBV treatment

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UO Epatologia – Azienda Ospedaliero Universitaria Pisana - Centro Riferimento Regionale *"Diagnosi e trattamento delle epatopatie croniche e del tumore di fegato"*

Disclosures

Advisory board: AbbVie, Gilead, Roche

Speakers' bureau: AbbVie, BMS, Gilead, MSD, Janssen

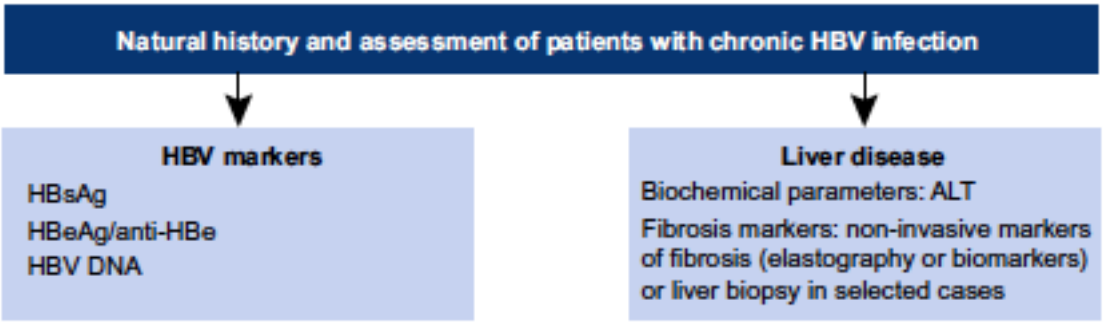
Research grant: AbbVie, BMS, MSD

EASL JOURNAL OF HEPATOLOGY

EASL 2017 Clinical Practice Guidelines on the management of hepatitis B virus infection☆

European Association for the Study of the Liver *

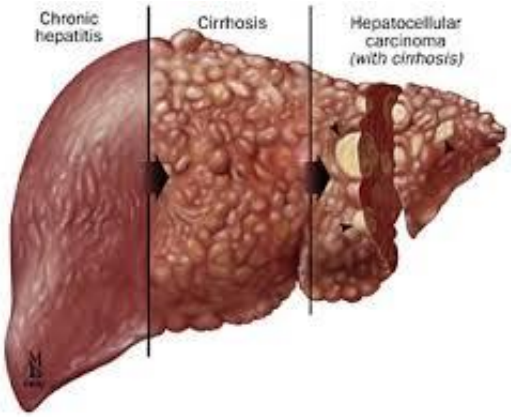
New nomenclature: based on the description of the 2 main characteristics of chronicity ➡ **infection** vs **hepatitis**



	HBeAg positive		HBeAg negative	
	Chronic infection	Chronic hepatitis	Chronic infection	Chronic hepatitis
HBsAg	High	High/intermediate	Low	Intermediate
HBeAg	Positive	Positive	Negative	Negative
HBV DNA	>10^7 IU/ml	10^4-10^7 IU/ml	<2,000 IU/ml°°	>2,000 IU/ml
ALT	Normal	Elevated	Normal	Elevated*
Liver disease	None/minimal	Moderate/severe	None	Moderate/severe
Old terminology	Immune tolerant	Immune reactive HBeAg positive	Inactive carrier	HBeAg negative chronic hepatitis

Cure vs Control

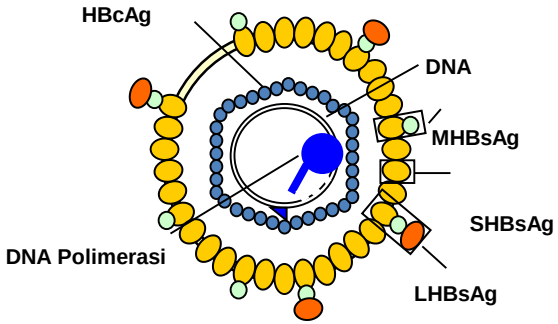
Liver disease



Cure: resolution of the chronic liver disease

Control: halting progression of the disease, without complete resolution, but with reduction/ elimination of the complications

HBV infection



Cure: clearance of the viral infection or complete shut-down of cccDNA

Control: achievement of an effective inhibition of viral replication, usually on therapy

Chronic Hepatitis B: from cure of the disease to its control

Cure **Control**

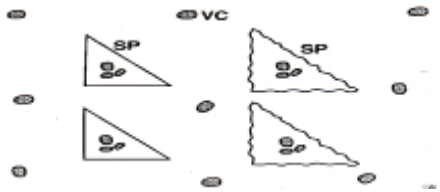
Portal
S 0-1

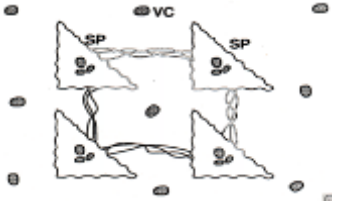
Periportal
S 2-3

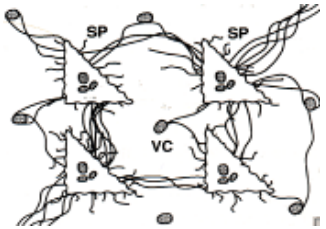
Septal
S 2-4

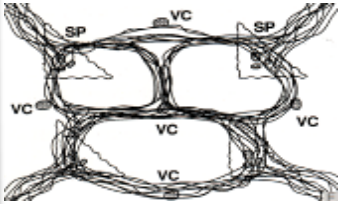
Inc. cirrhosis
S 3-5

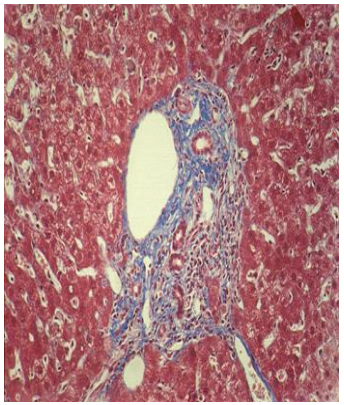
Comp.Cirrhosis S
4-6

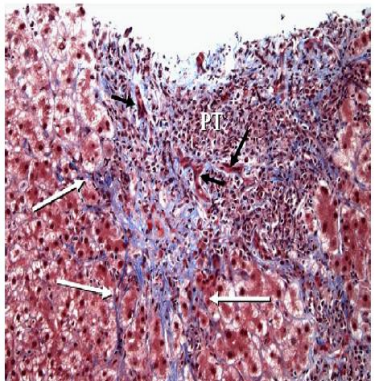


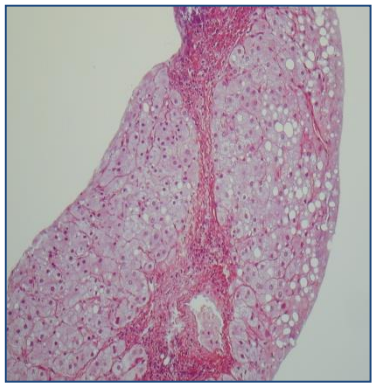


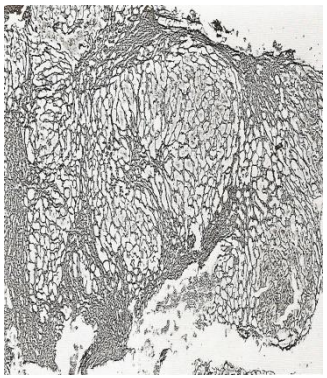


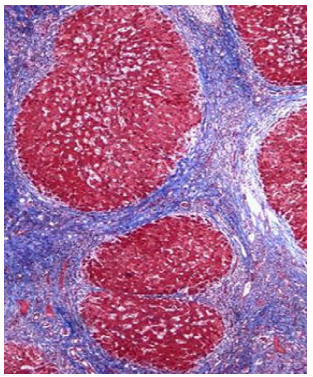












1- 6 yrs

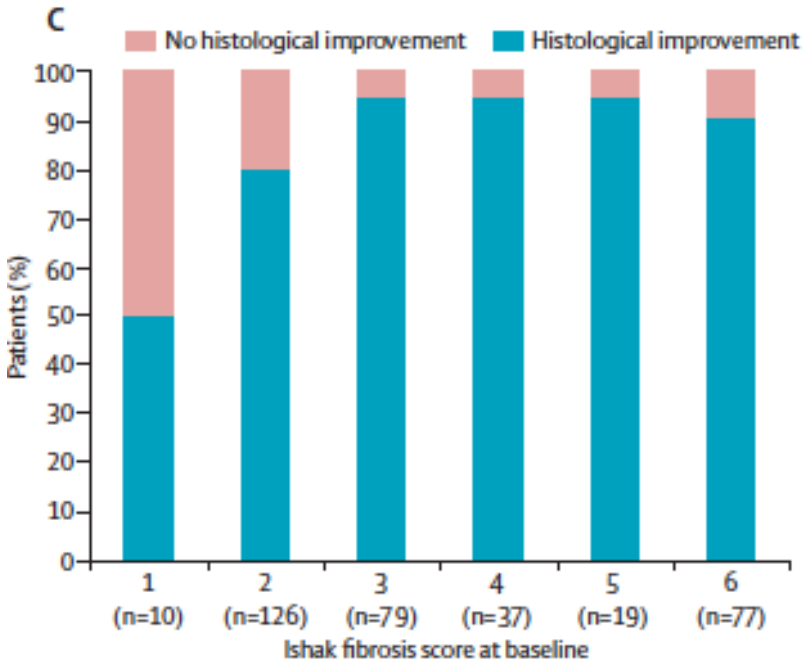
5 - 15 yrs

5 - 15 yrs

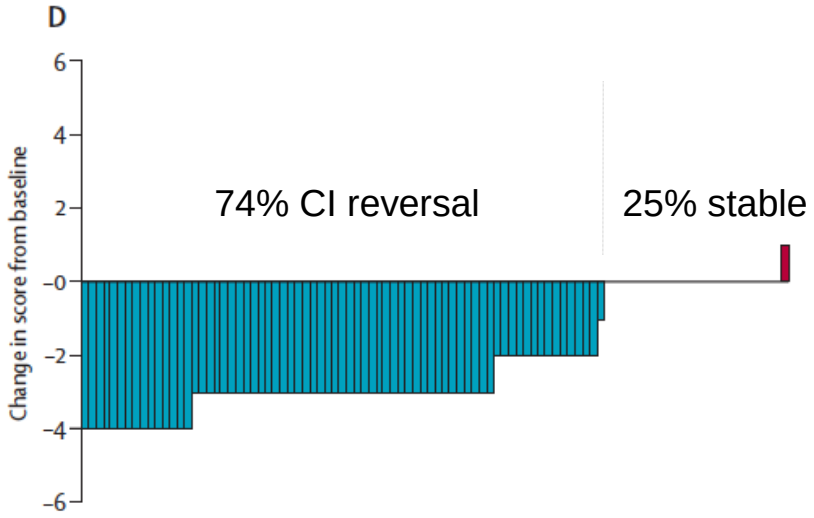
1 - 6 yrs

Regression of cirrhosis during treatment with TDF for CHB: a 5 year open label follow-up study

- Of 641 patietns who received randomized treatment, 585 entered the open label phase and 489 (76%) completed 240 weeks: **348 pts (54%) had liver biopsy at BL and 240 w.**
- **87%** (304) of the pts had **histological improvement** and **51%** (176) **regression of fibrosis** at w 240.



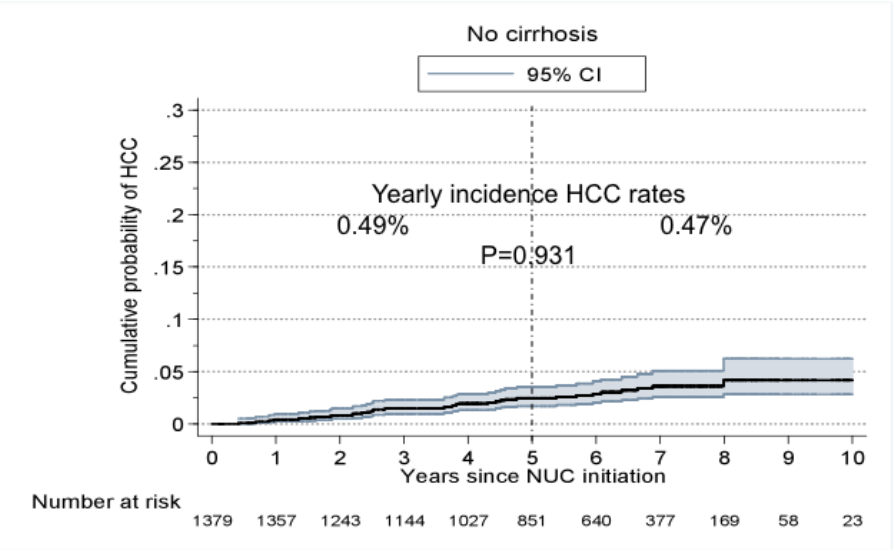
Change in Ishak score in 96 cirrhotics

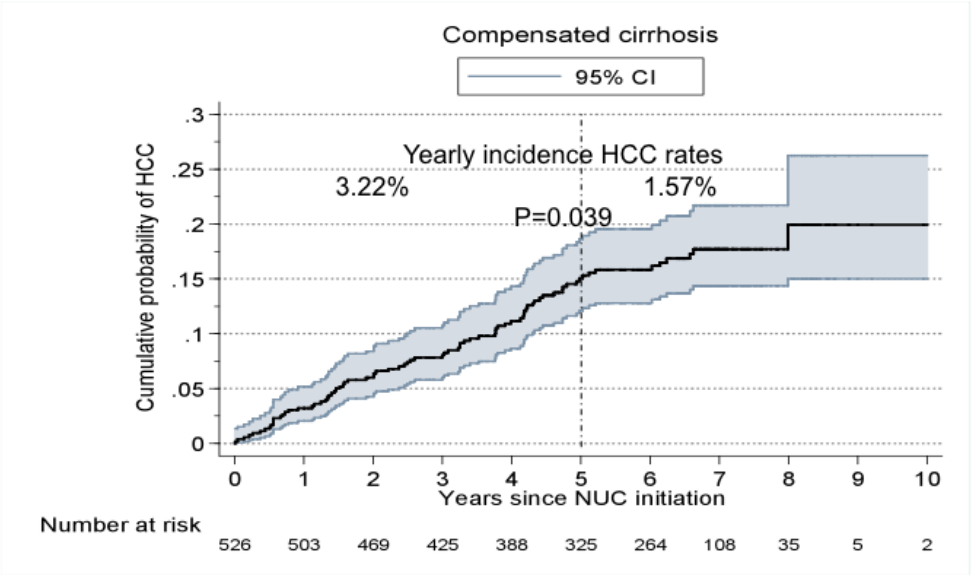


BMI, history of diabetes, abnormal ALT and Knodell score >3 were associated with persistence of cirrhosis at 5 y.

Marcellin P, Lancet 2013

The PAGE B cohort study: 10-year cumulative probability of HCC during NUC therapy



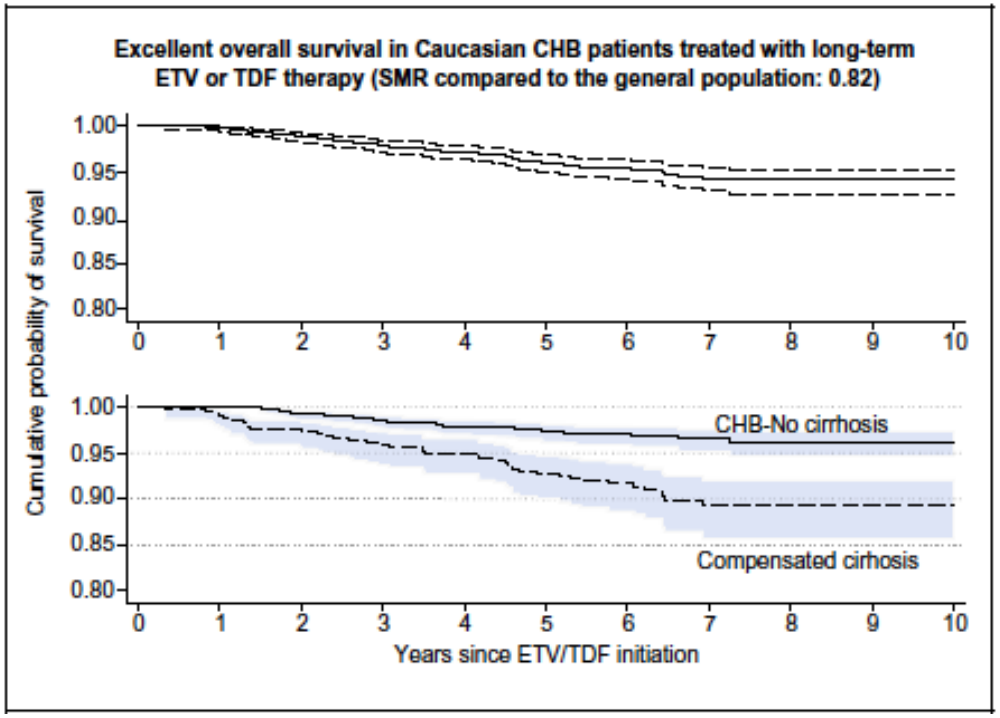


After the first 5 years of ETV or TDF therapy in CHB patients, the HCC incidence is decreasing, with the decrease being more evident in patients with baseline cirrhosis

Papatheodoridis G et al, Hepatology 2017

Eight-year survival in CHB patients under long-term ETV and TDF therapy is similar to the general population

1951 adult Caucasian with CHB with (27%) /without cirrhosis, without HCC at BL on NUCs for at least 12 months /median 6 years)



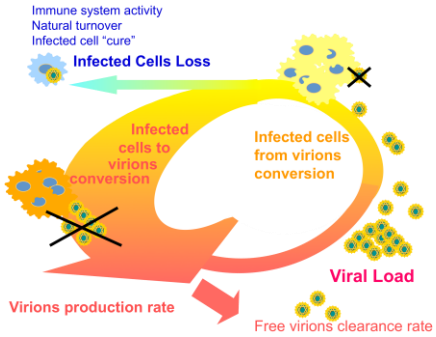
	Survival propability at			
	1 y.	3 y.	5 y.	8 y.
CHB without CI	100	98.5	97.3	96.2
CHB with CI	99.1	95.9	92.8	89.3

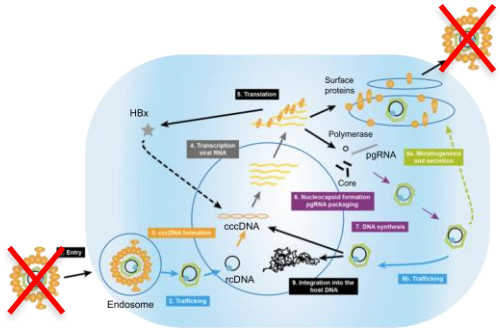
- Mortality of non-cirrhotics seems to be lower than that of the general population
- Mortality of cirrhotics is similar to that of the general population
- HCC is the main factor affecting patient mortality
- 2.6% of the patients had major liver related events events (death od OLT)

Overall mortality rate was 0.77 (95%CI 0.62-0.96) comparable to that of the general population, 0.82 (95%CI 0.66-1.039)

Papatheodoridis G et al, J Hep 2018

Response to antiviral treatment: how it is changing the way to look at it





Virological responses

- *during NA* **undetectable HBV DNA** by a sensitive PCR (LoD 10 IU/ml)
- *after NA*, sustained off-therapy virological response, **HBV DNA <2,000 IU/ml** for at least 12 months
- *during PegIFNa* **HBV DNA <2,000 IU/ml** at 6 months and at the end of therapy.
- *after PegIFNa* **HBV DNA <2,000 IU/ml** for at least12 months

Serological responses

- **HBeAg** are HBeAg loss and HBeAg seroconversion
- **HBsAg** are HBsAg loss and HBsAg seroconversion

Complete sterilising cure:

undetectable serum **HBsAg** and **eradication of HBV-DNA** including intrahepatic cccDNA and integrated HBV-DNA

Functional cure:

sustained, **undetectable** serum **HBsAg** and **HBV-DNA** with/without seroconversion to anti-HBs *(several levels of functional cure according to cccDNA status: complete shut down of cccDNA transcription or its elimination)*

Partial cure:

detectable serum HBsAg, but **persistently undetectable** serum **HBV-DNA**

EASL, HBV CPG 2017

Lok A et al, J Hep 2018

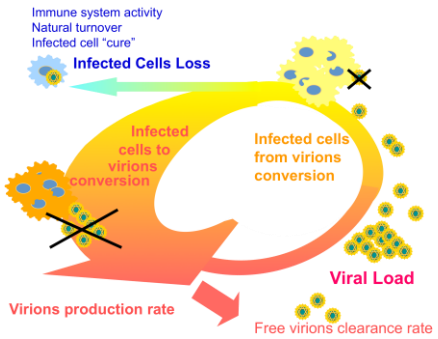
Why to cure HBV infection?

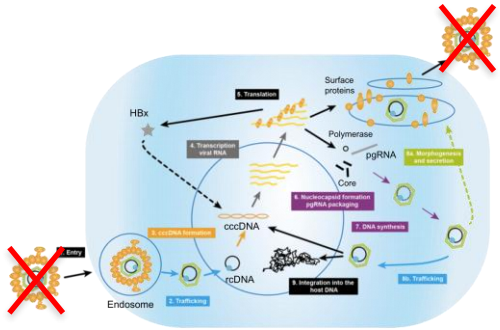
- To achieve a persistent control of HBV infection off therapy in a higher proportion of cases
- To achieve a complete inhibiton of HBV replication
- To halt events that may have oncogenetic potential

- To increase the therapeutic index of antiviral treatment
- To increase the number of patients who could have cured their chronic hepatitis B

Magritte, Decalcomania

Response to antiviral treatment: how it is changing the way to look at it





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- **HBsAg** are HBsAg loss and HBsAg seroconversion

On or off therapy

EASL, HBV CPG 2017

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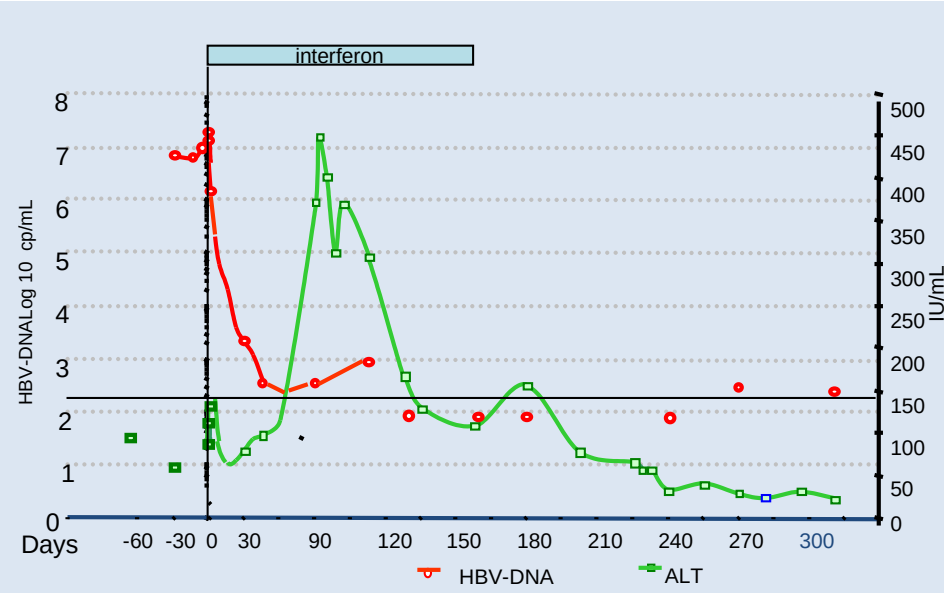
Partial cure:

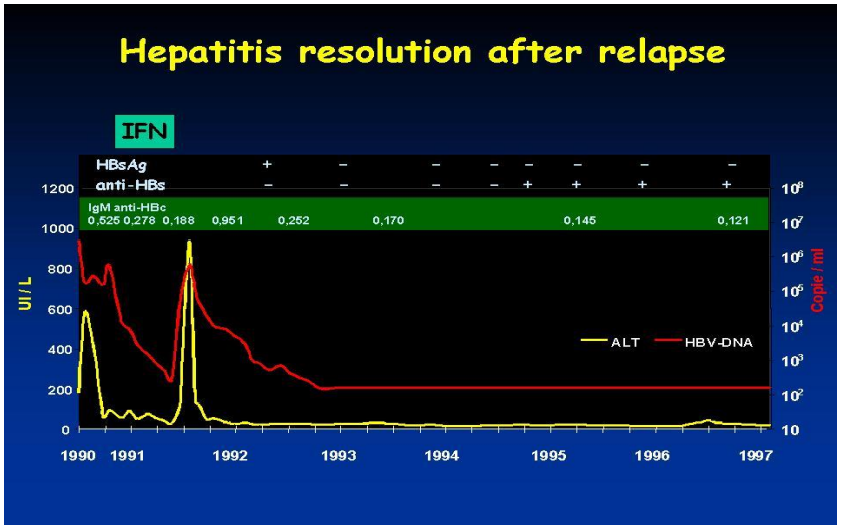
detectable serum HBsAg, but **persistently undetectable** serum **HBV-DNA**

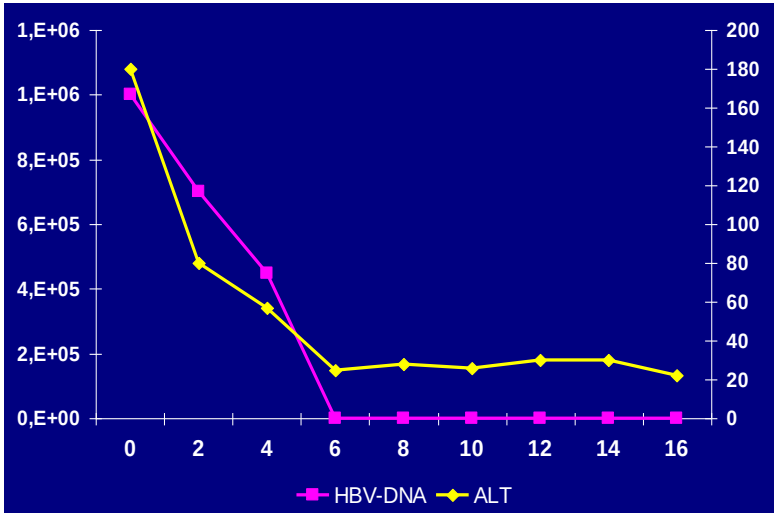
Off therapy

Lok A et al, J Hep 2018

Successful antiviral treatment in CHB patients





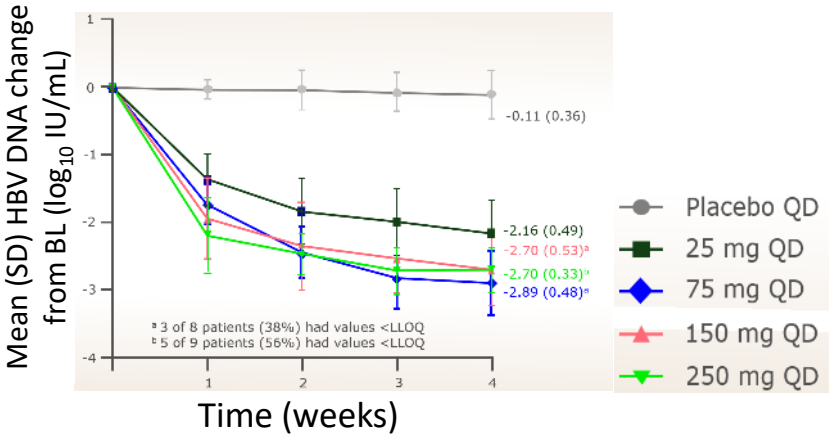


Brunetto MR et al, Hepatology 1989

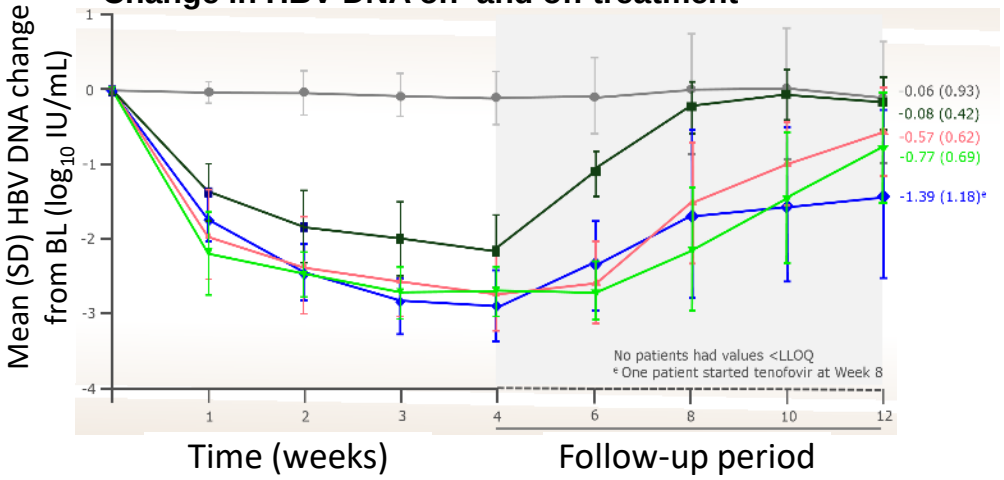
Phase 1 studies with new drugs for the treatment of CHB

HBV capsid assembly modulator, JNJ-56136379

Change in HBV DNA on treatment

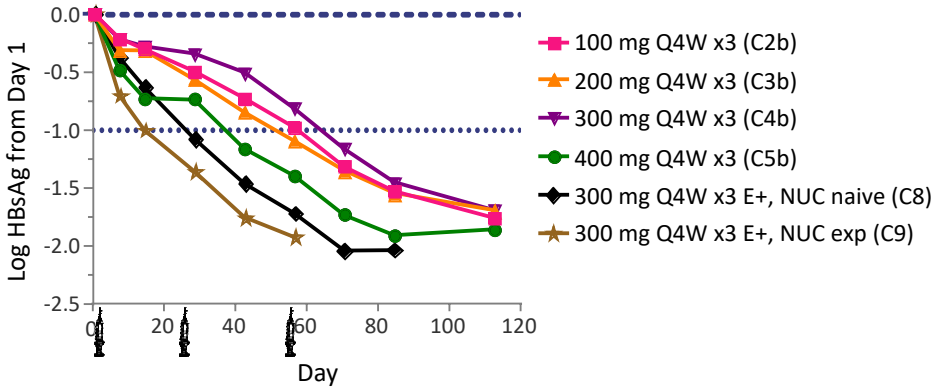


Change in HBV DNA on and off treatment



RNA interference (RNAi), ARO-HBV

Change in HBsAg from day 1



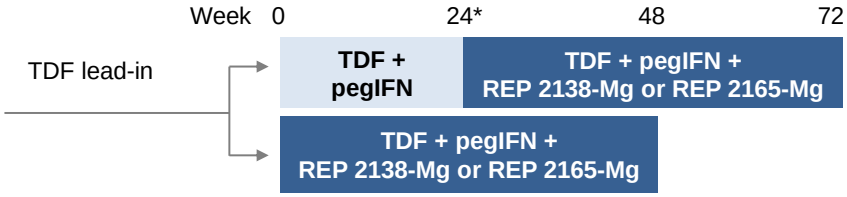
Phase 1 studies are mainly aimed to test:

- Safety
- Tolerability
- Pharmacokinetics
- Antiviral activity

Zoulim F, et al. AASLD 2018, San Francisco, USA. #74

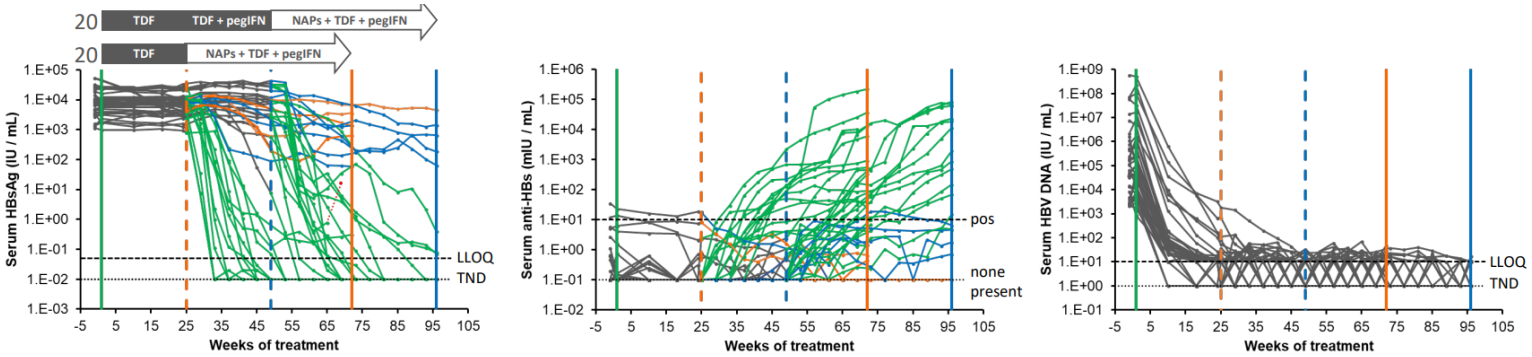
Gane E, et al. AASLD 2018, San Francisco, USA. #LB-25

REP 2139-Mg / REP 2165-Mg, TDF and pegIFNα-2a in patients with HBeAg negative CHB

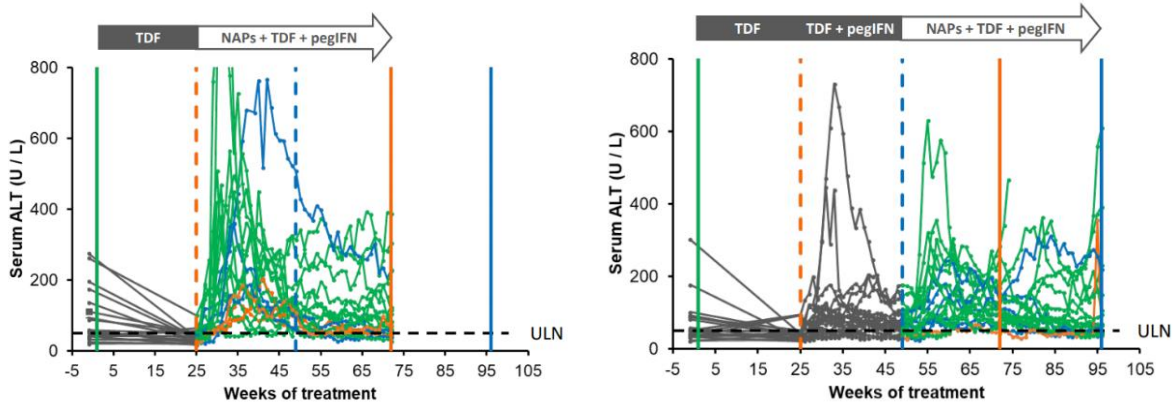


*Crossed over at 24 wks due to poor HBsAg response (< 3 log reduction in HBsAg)

- 40 patients
- 5 with advanced fibrosis (according to Fibroscan values)



Virological response



ALT profile

38/40 pts had ALT flare concomitantly with REPs exposure

ALT flare ?

bad

good

At present the definition is **only** based on the ***ex post*** evidence of

Good, beneficial, therapeutic flares

- when they are associated with the achievement of the control of the infection

Virologic response

- because asymptomatic
- because self-limited
- because not associated with liver dysfunction

Absence of clinically relevant events

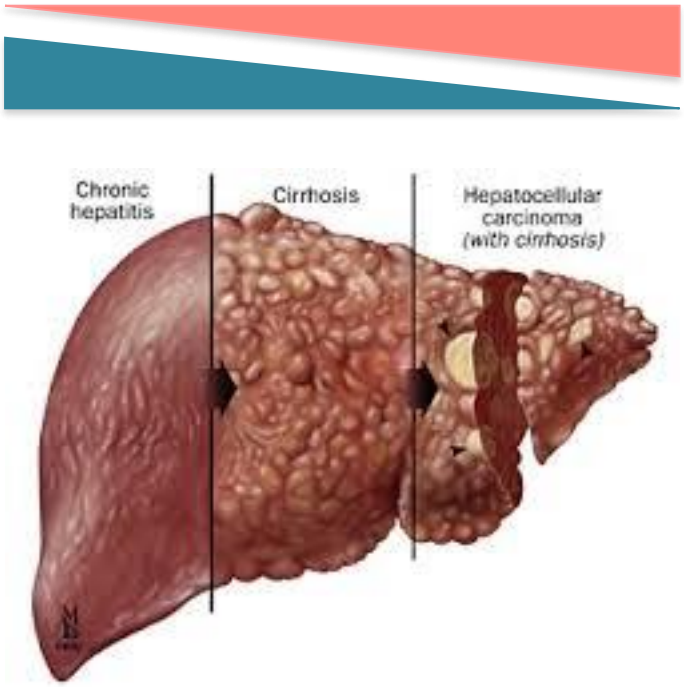
However, ***a priori*** the flare remains **always** potentially threatening in a patient with advanced fibrosis and reliable predictive factors are missing

Control or Cure of HBV infection for a personalized treatment

Type of drugs' combination according to:

- ✓ Virus and infecting viral population
- ✓ Phase of HBV infection
- ✓ Stage of liver disease
- ✓ Treatment status (naive or on treatment)
- ✓ Host features (age, gender, co-morbidities)

HBV cure vs
HBV infection control



Liver disease control

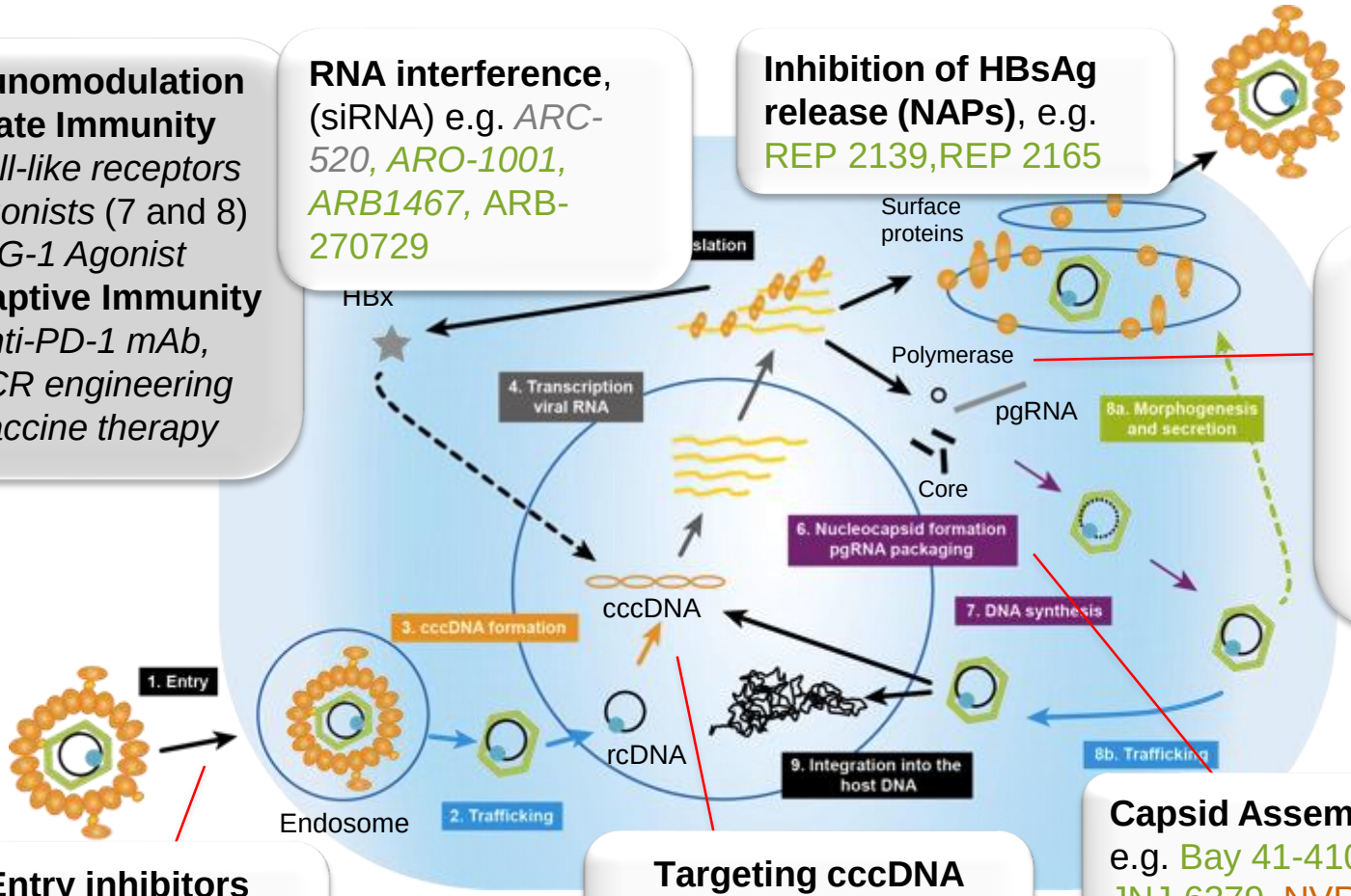
Antifibrotic, antiangiogenetic drugs

Future HBV therapies: new targets, new drugs

Immunomodulation
- **Innate Immunity**
  Toll-like receptors agonists (7 and 8)
  RIG-1 Agonist
- **Adaptive Immunity**
  Anti-PD-1 mAb,
  TCR engineering
  Vaccine therapy

RNA interference, (siRNA) e.g. *ARC-520, ARO-1001, ARB1467,* ARB-270729

Inhibition of HBsAg release (NAPs), e.g. REP 2139,REP 2165



Polymerase inhibitors
- Nucleoside analogues, e.g.
- TAF, amdoxovir, MIV-210
- Non-nucleoside, e.g. LB80380

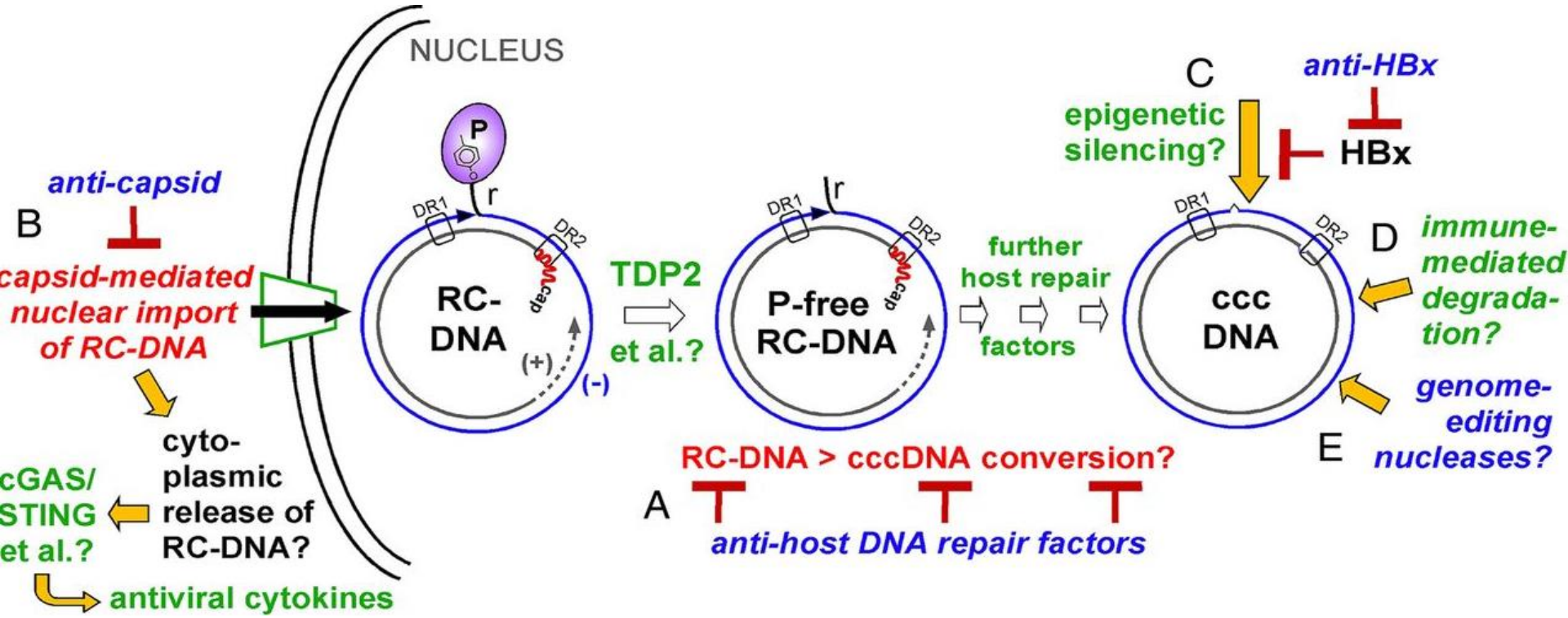
Capsid Assembly Modulators, e.g. Bay 41-4109, RO7049389, JNJ-6379, NVR1221, AB-506

Targeting cccDNA
- HAPs
- Chromatin-modifying enzymes

Entry inhibitors (HBV/HDV)
- Lipopeptides, e.g. Myrcludex-B

Development stage: **preclinical**, **clinical** ; modified and updated from Zoulim F, et al. Antiviral Res 2012;96(2):256–9; HBV Drug Watch, Available at: http://www.hepb.org/professionals/hbf_drug_watch.htm.

HBV Direct Antivirals: Targeting cccDNA

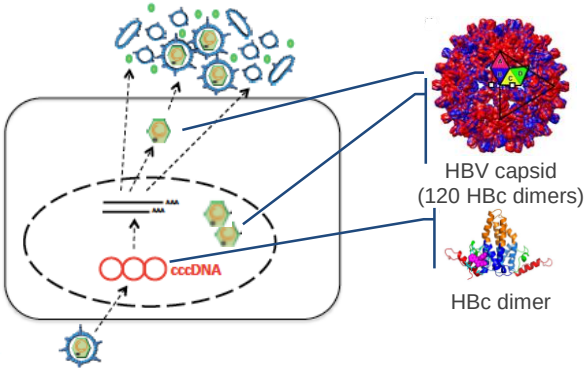


Strategies targeting the cccDNA:

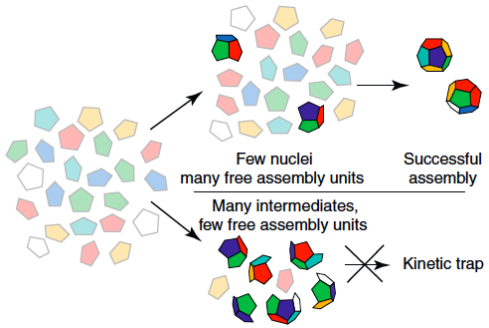
- cccDNA formation
- eliminating existing cccDNA
- silencing cccDNA transcription

Nassal et al, Gut 2015

Targeting HBc protein / HBV capsid by Core inhibitors / Core Protein Assembly Modulators (CpAM)



- ❖ Capsid formation and pgRNA encapsidation pivotal for infective particles production
- ❖ HBc binds the cccDNA and modifies cccDNA nucleosome spacing



Capsid Assembly Modulators (CpAM):

- class 1 CpAM (RO7049389) induces abberrant core protein aggregates, that are subsequently degradated
- class 2 CpAM (JNJ-6379) block pgRNA encapsidation with production of empty capsids

Gane E, AASLD 2014

RO7049389, a core protein allosteric modulator (CpAM), is effective against HBV, safe and well tolerated

RO7049389 is an oral small molecule, Class I HBV CpAM

Methods:

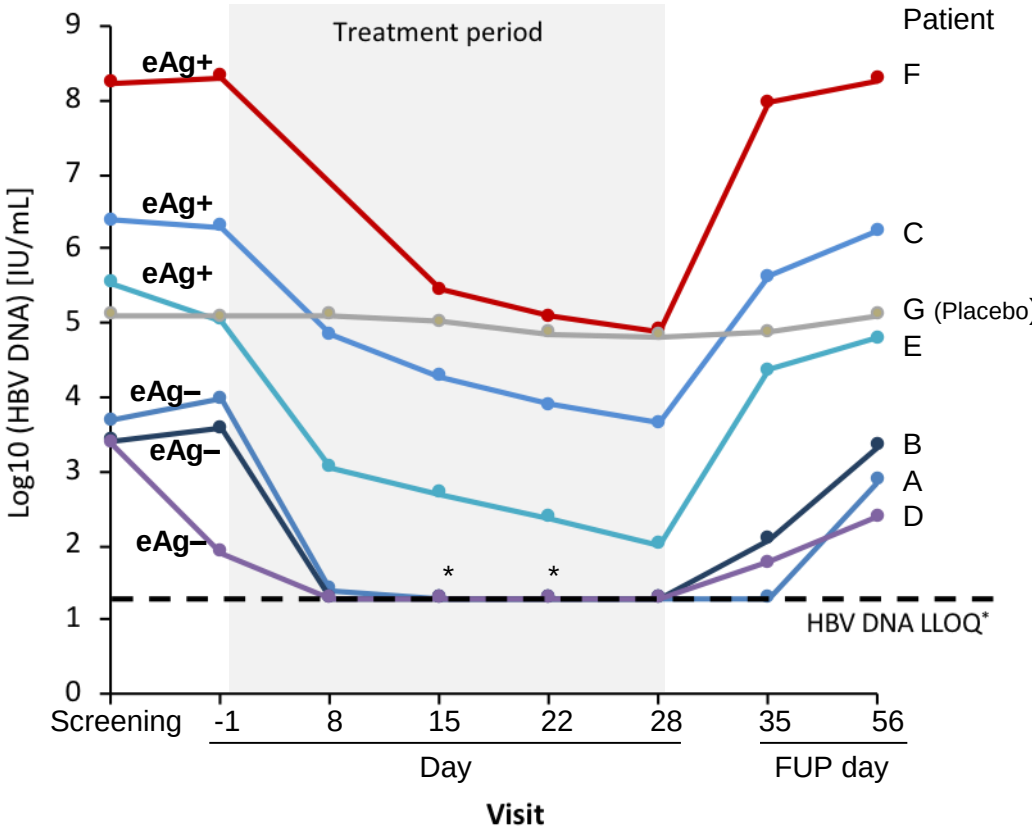
YP39364 is a Phase I study investigating safety, PK and anti-viral activity of RO7049389

- SAD and MAD cohorts in healthy volunteers have been completed
- Dosing in the first patient cohort at 200mg bid for 28 days is complete

Results:

- To date, RO7049389 is well tolerated with no clinically significant changes or trends evident in safety data
- Robust HBV DNA declines: median (maximal) –2.7 (–3.4) log_{10} IU/ml, observed to Day 28 of dosing 200 mg bid (n=6)
- Future cohorts will explore higher doses and once daily dosing

Absolute HBV DNA change over time



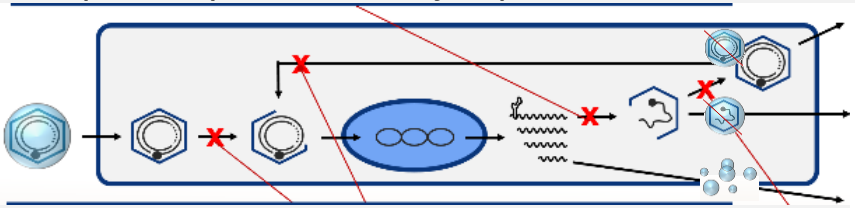
*HBV DNA Values <LLOQ and below the lower limit of detection

Gane E, et al. ILC 2018, #LBO-003

Safety, PK and antiviral activity of a novel HBV capsid assembly modulator, JNJ-56136379, in patients with CHB

JNJ-6379 is a CAM that binds to HBV core protein and disrupts early and late-stage processes in the HBV lifecycle

"Primary" mechanism ("empty capsid" CAM)
Interference with capsid assembly kinetics, preventing encapsidation of (pg)RNA and blocking HBV replication (late step in viral life cycle) - **JNJ-6379 median EC_{50}/EC_{90} = 102/376 nM**



Dane particle (infectious DNA containing)

RNA-containing particle (pgRNA, spliced RNA)

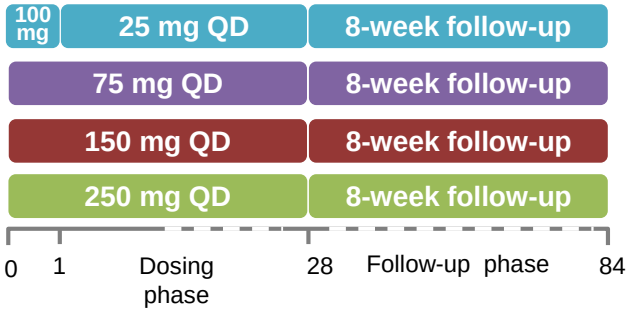
Subviral particles (HBsAg)

"Secondary" mechanism
Inhibition of de-novo formation of cccDNA, potentially by interfering with capsid disassembly process (early stage in viral life cycle) - **JNJ-6379 median EC_{50}/EC_{90} = 876/4019 nM**

NAs block HBV replication but do not inhibit production of RNA-containing particles

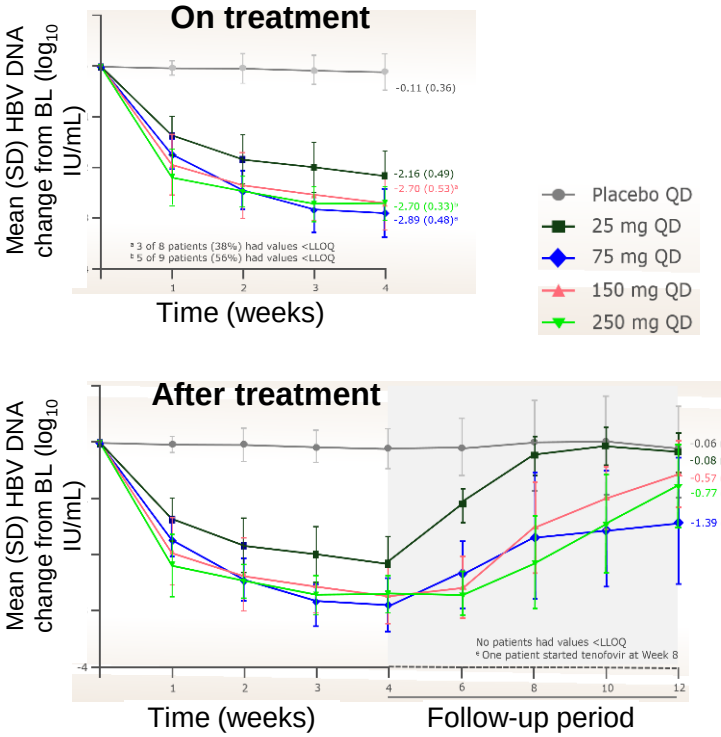
- HBeAg +ve or –ve
- Non-cirrhotics (F0–F2)
- Plasma HBV DNA >2000 IU/mL
- ALT <2.5 x ULN

Session 8 (EU) (8 drug; 4 PBO)
Session 9 (EU) (8 drug; 4 PBO)
Session 10 (EU/AP) (9 drug; 3 PBO)
Session 11 (EU/AP) (9 drug; 3 PBO)

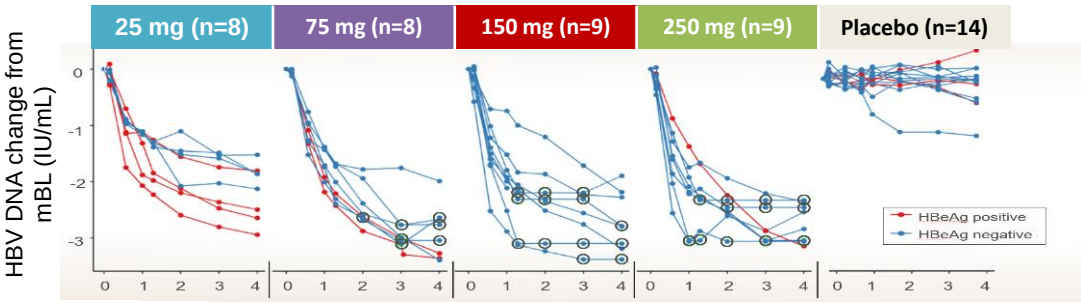


Safety, PK and antiviral activity of a novel HBV capsid assembly modulator, JNJ-56136379, in patients with CHB

- **Change in HBV DNA**



HBV DNA in individual patients



Mean HBV RNA change from baseline after 4 weeks on treatment

Treatment arms	Baseline[a]	End of Week 4	
	Mean (SD) log_{10}copies/mL	Mean (SD) change from BL log_{10}copies/mL	Not detected, n (%)
25 mg (n=8)	5.59 (2.37)	–2.30 (0.59)	3 (38)
75 mg (n=8)	3.39 (2.21)	–1.85 (1.42)	6 (75)
150 mg (n=9)	3.37 (1.66)	–1.83 (0.93)[b]	6 (75)[b]
250 mg (n=9)	2.58 (1.94)	–1.43 (1.13)	8 (89)
Placebo (n=14)	2.92 (2.46)	0.02 (1.10)	5 (36)

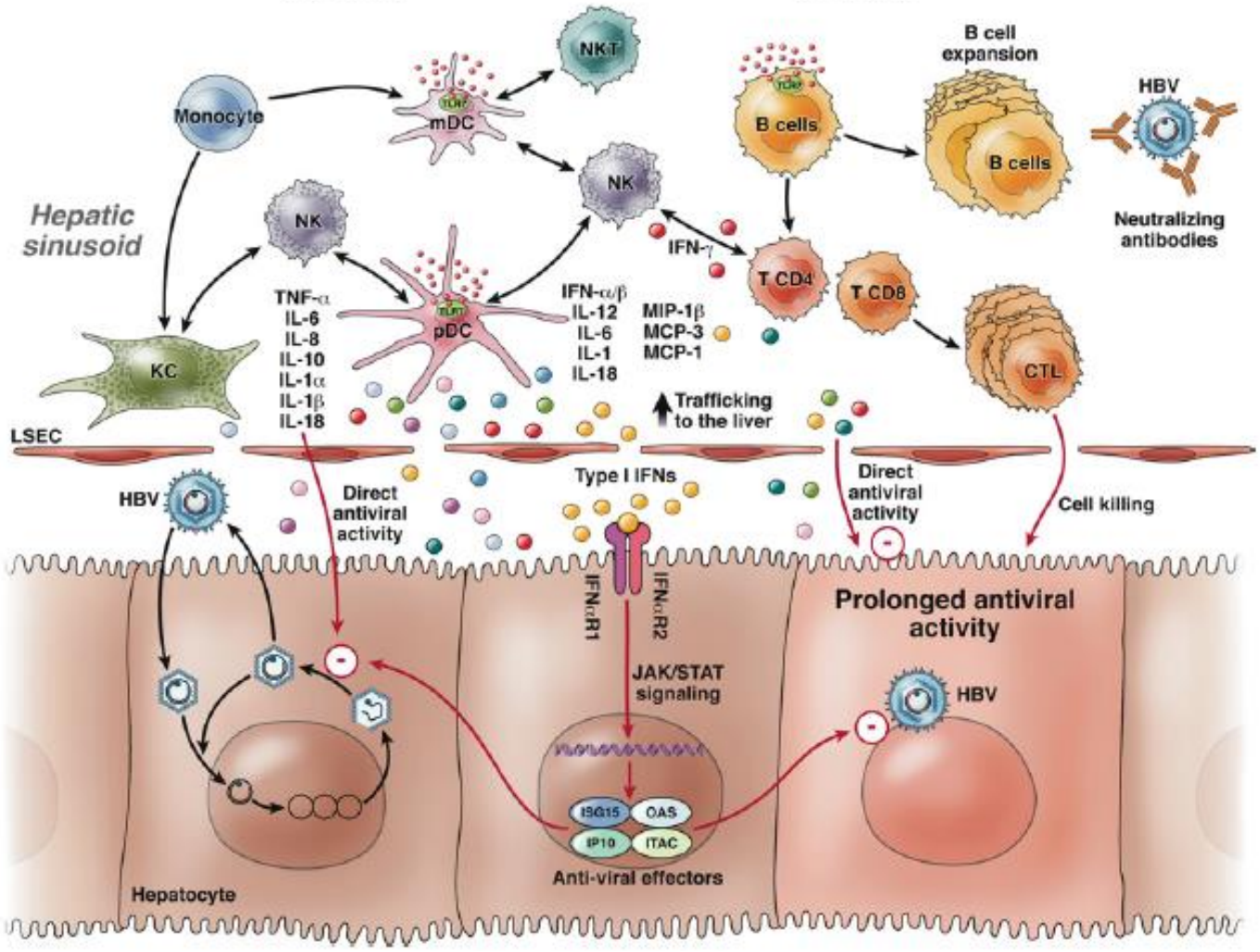
- Dose-related suppression of viral replication, majority of high-dose patients became HBV DNA <LLQ
- No reduction in HBsAg but treatment duration short
- One on-treatment Grade 4 ALT elevation

Poor innate immune response with limited quantity of cytokines (IFN-α, IL-6, TNF-α)

HBV specific T-cell dysfunction, due to persistent exposure to high Antigen doses, T-reg suppression, Dendritic cell impairment

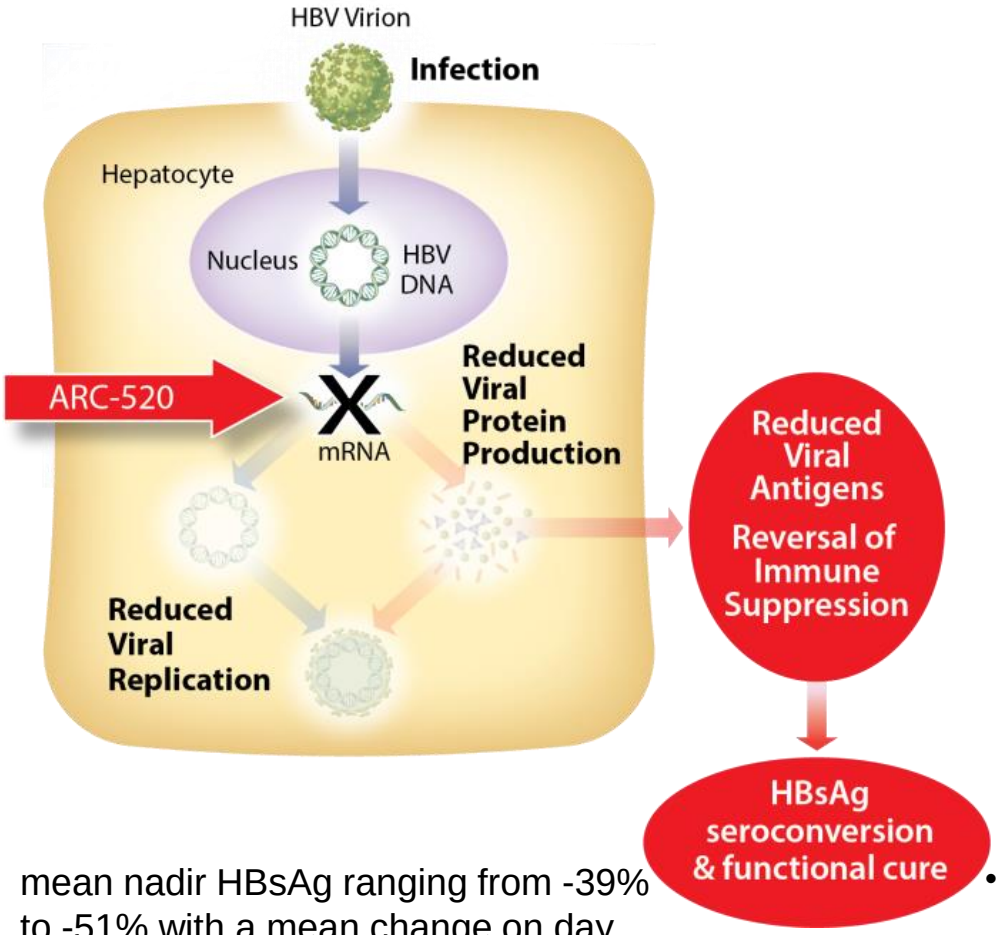
Innate Immunity

Adaptive Immunity



S.Wieland et al PNAS 2004; A.Stacey et al. J. Virology 2009,; Dunn et al. Gastroent. 2009; Zoulim F et al. Gastroent. 2014

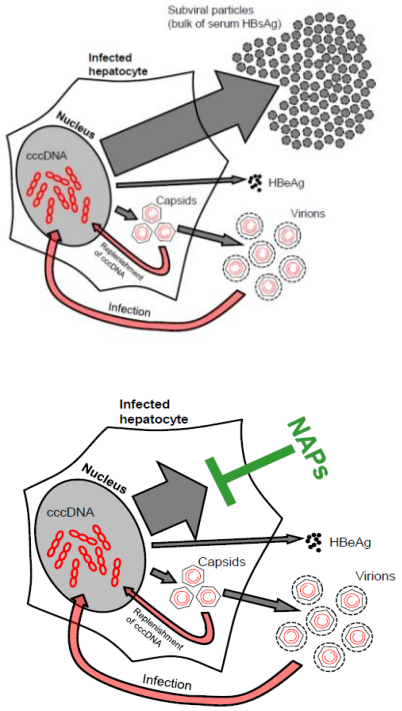
Inhibition of HBsAg production



- mean nadir HBsAg ranging from -39% to -51% with a mean change on day 85 of 31-22%
- This is the first time that a reduction in HBsAg mediated through RNA interference has been shown in chronic HBV patients.

Yuen MF et al, AASLD 2014

Nucleic Acid Polymers (NAPs)

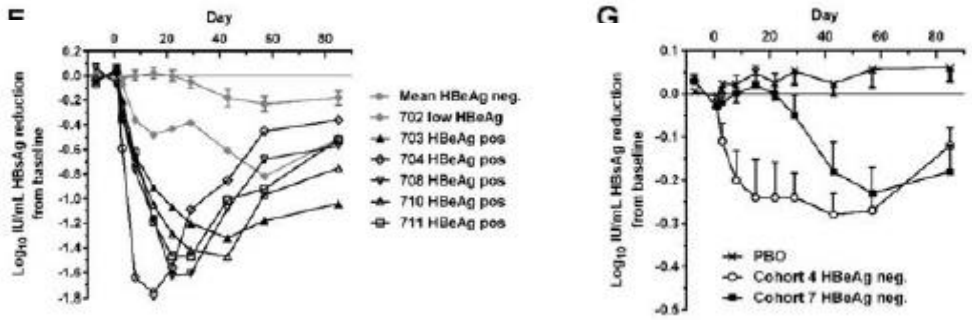


- REP 9-AC HBsAg serum clearance in 7/8 HBeAg + pts with durable HBV-DNA reduction in 2/7 responders
- REP 2139 monotherapy, followe by combination with Peg-IFN and Peg-IFN monotherapy led to undetectable HDV-RNA in 10/12 HDV pts, without rebound of HBsAg in pts with lelev < 10 IU/ml

Mahtab MA et al Hepatology 2011, Bazinet et al J Hep 2016

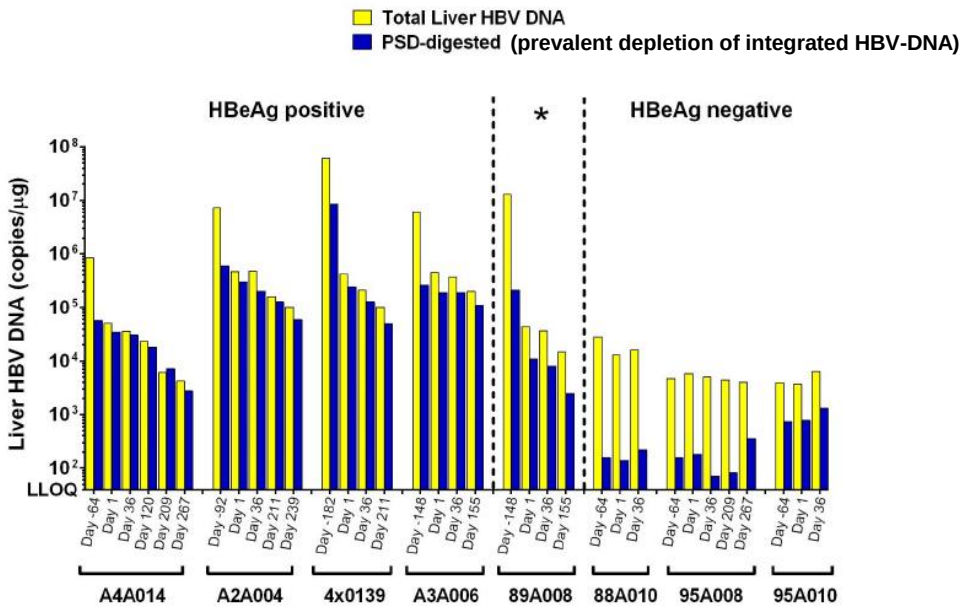
RNAi-Based Treatment of Chronically Infected Patients and Chimpanzees Implicates Integrated Hepatitis B Virus DNA as a Source of HBsAg

- **siRNA in ARC-520 was designed to target all HBV mRNAs**



ARC-520 reduced significantly serum HBsAg and HBeAg in **HBeAg positive naive** pts, **but not** in **HBeAg negative** pts or **HBeAg pos on** prolonged **NUC** treatment

Quantification of liver HBV-DNA in ARC-520 treated chimps



- **ARC-520 was not active mRNAs from integrated HBV-DNA**

- Studies in chimps suggest a dramatic increase of the integration in the HBeAg negative phase.
- >90% of the mRNAs deriving from integrated HBV-DNA

- A significant proprotion of HBsAg in HBeAg negative patients could derive from integrated HBV sequences

Wooddell C et al Sci Transl Med 2017

First results with RNA interference (RNAi) in CHB using ARO-HBV

AROHBV1001 background

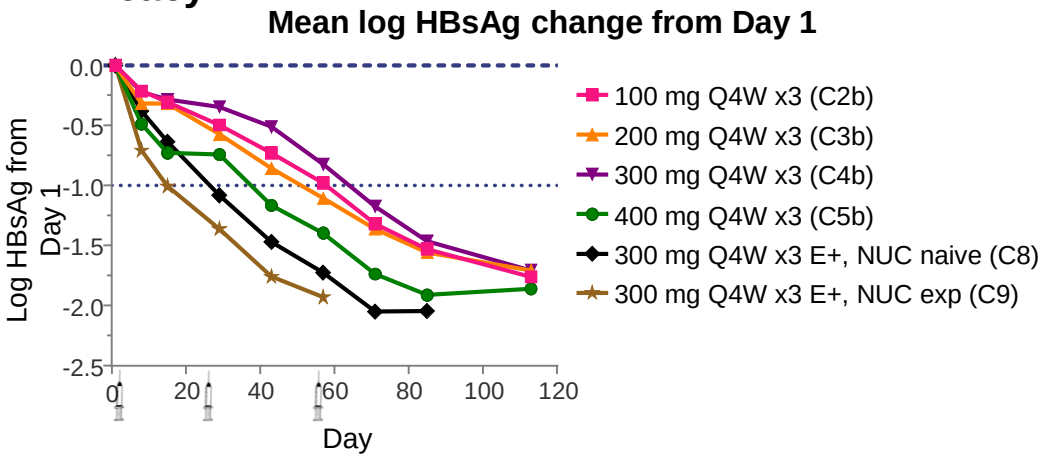
- ARC520/521 RNAi program demonstrated need to target integrated HBs in HBeAg –ve CHB patients
- Membrane lipid protein delivery - endos required IV administration, associated with preclinical toxicity
- ARO-HBV is new TRiM™ delivery system for RNAi

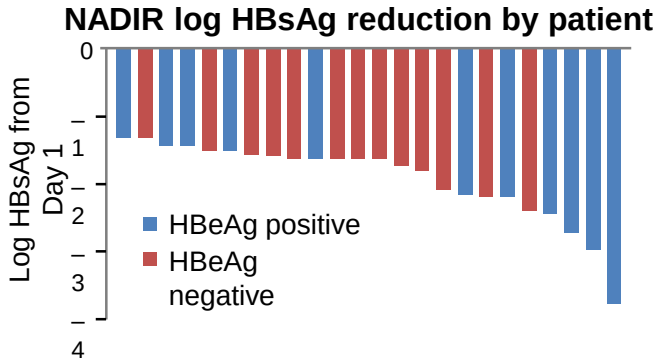
- **X and S triggers → silence all mRNA from both cccDNA and host integrated viral DNA**
- **GAL-NAC conjugation allows SC administration**

AROHBV1001 study design

- SAD in 6 cohorts of healthy volunteers (4 active: 2 PBO) received 35, 100, 200, 300, 400 mg
- MAD in 6 cohorts of CHB patients (4, open-label) received ARO-B 100, 200, 300, 400 mg Q4W x3
  - Cohorts 2b–5b HBeAg +/–ve NUC-naive or suppressed
  - Cohort 8 HBeAg pos naïve (start NUC on Day 1)
  - Cohort 9 HBeAg+ve suppressed
- **Endpoints:** PK, safety and tolerability of ARO-HBV
  - HBsAg reduction from baseline in CHB patients

Efficacy

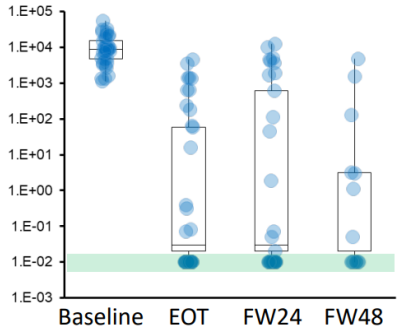




- ARO-HBV well tolerated at single or multiple monthly SC doses up to 400 mg
- Robust HBsAg decline in all HBV patients (1.3–3.8 $\log_{10}$) – slower than with IV ARC-520/521
- No clear dose-response above 100 mg
- **HBsAg declines similar in HBeAg +ve and HBeAg –ve**
- **Associated DNA, RNA, HBeAg, HBcrAg responses**

High rates of functional control and reversal of fibrosis following treatment of HBeAg negative chronic HBV infection with REP 2139-Mg / REP 2165-Mg, TDF and pegIFNα-2a

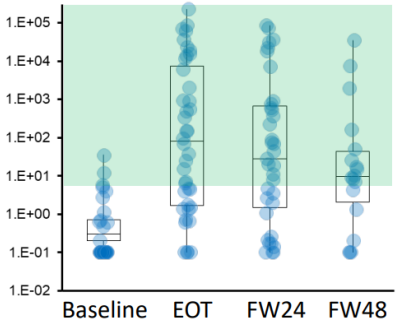
HBsAg (IU/mL)



0% (0/40) | 60% (24/40*) | 53% (18/34) | 50% (8/16)

HBsAg loss (≤ 0.05 IU/mL)

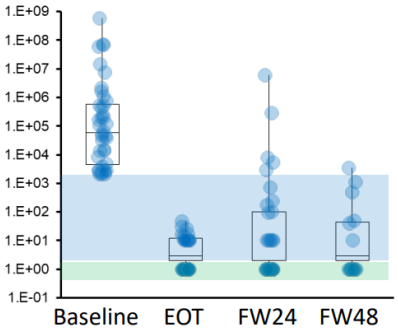
Anti-HBs (mIU/mL)



5% (2/40) | 60% (24/40*) | 59% (20/34) | 56% (9/16)

HBsAg seroconversion (Anti-HBs ≥ 10 mIU/mL)

HBV DNA (IU/mL)



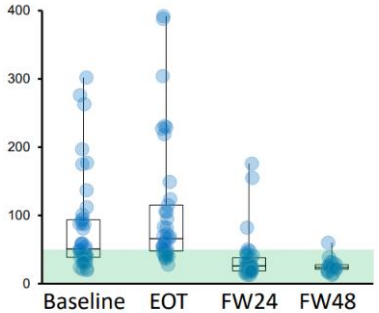
0% (0/40) | 55% (22/40*) | 50% (17/34) | 62% (10/16)

HBV DNA target not detected

0% (0/40) | 45% (18/40*) | 38% (13/34) | 31% (5/16)

HBV DNA BLOQ to 2000 IU/mL

ALT (U/mL)



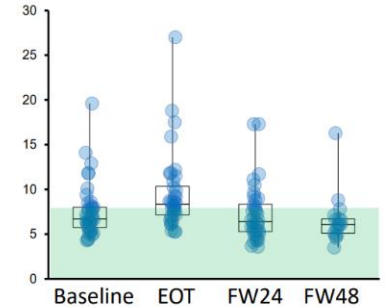
47% (19/40) | 32% (13/40*) | 91% (31/34) | 94% (15/16)

Normal ALT (≤ 50 U/L)

- 3 patients withdrew from therapy early for personal reasons

BLOQ; below the lower limit of quantification

Major hepatic stiffness (kPa)



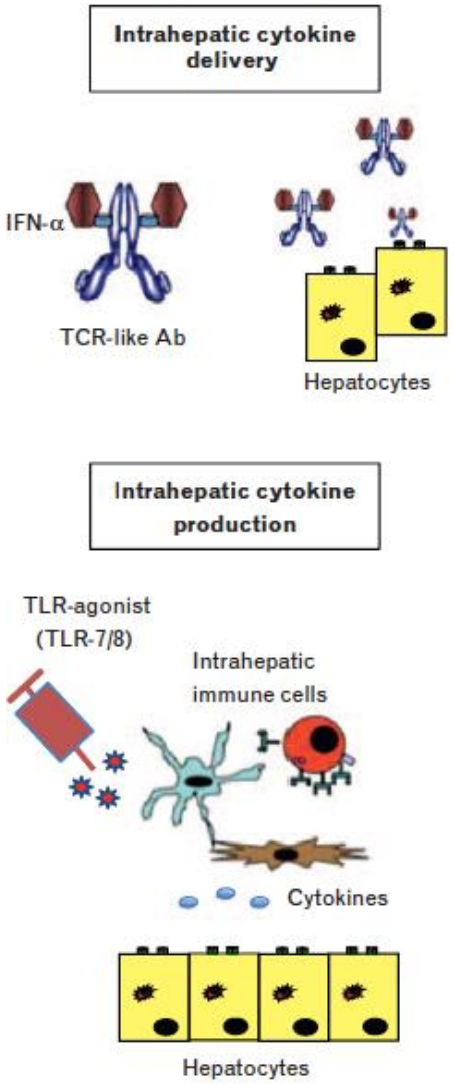
52% (21/40) | 22% (9/40*) | 62% (21/34) | 81% (13/16**)

Median hepatic stiffness consistent with F0 (≤ 7 kPa)

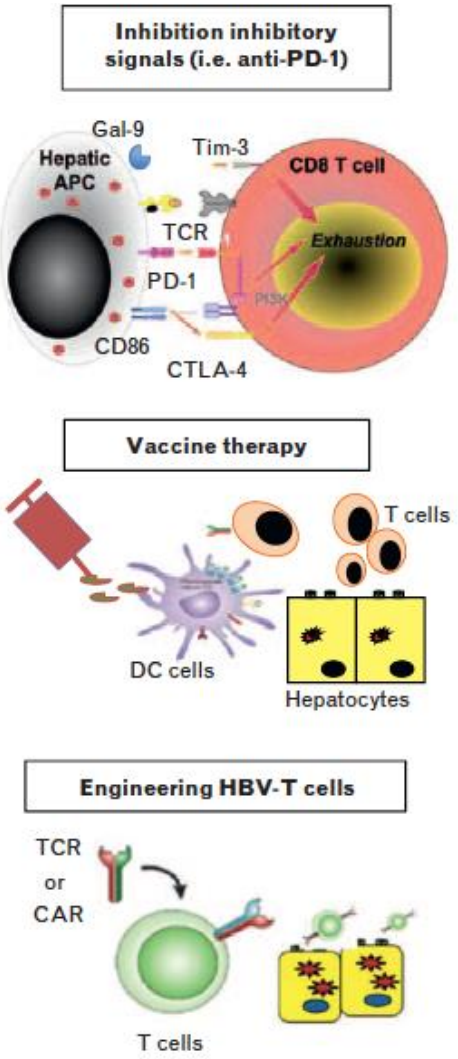
- ALT flares were observed in 38/40 patients concomitantly with REP 2139-Mg / REP 2165-Mg exposure.
- ALT flares were self-resolving, otherwise asymptomatic and correlated with reductions in serum HBsAg during treatment.
- 44% of the patients at 24-48 post-treatment follow-up show the HBeAg negative infection profile and 41% a functional cure
- REP 2139-Mg/REP 2165-Mg are effective and well tolerated in combination with pegIFN and TDF in HBeAg neg. CHB and elicit the establishment of functional control of HBV infection persisting after removal of therapy.

Bazinet M, et al. AASLD 2018, San Francisco, USA. #393

Boosting innate immunity



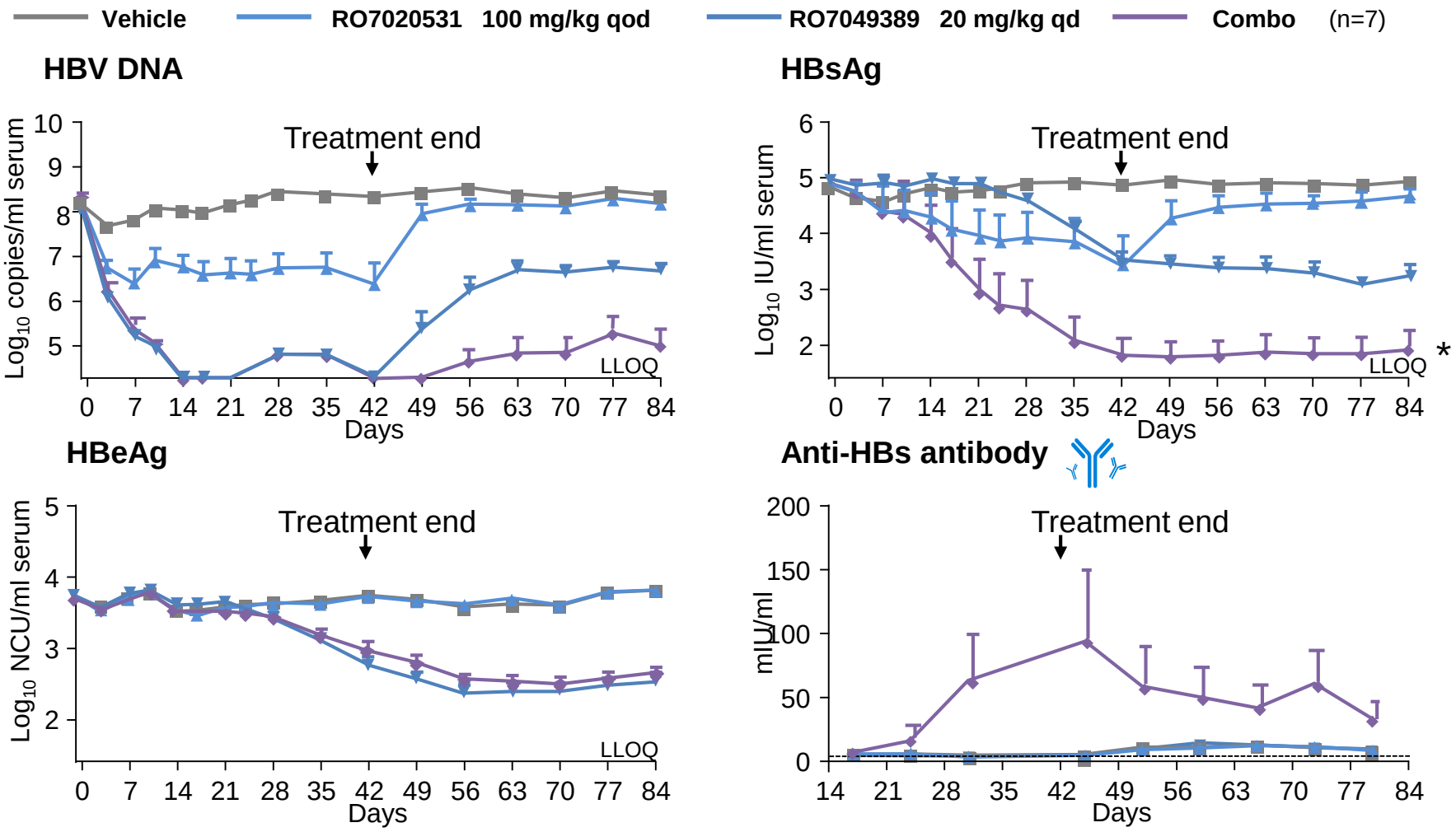
Boosting HBV specific Adaptive immunity



- To avoid unspecific boosting of innate immunity
- To tailor the HBV specific immunity limiting the killing of infected hepatocytes

Adapted from Bertoletti A et al, Curr Op Infect Dis 2014

TLR7 agonist RO7020531 + CpAM RO7049389 achieved sustainable VL suppression and HBsAg loss in an AAV-HBV mouse model



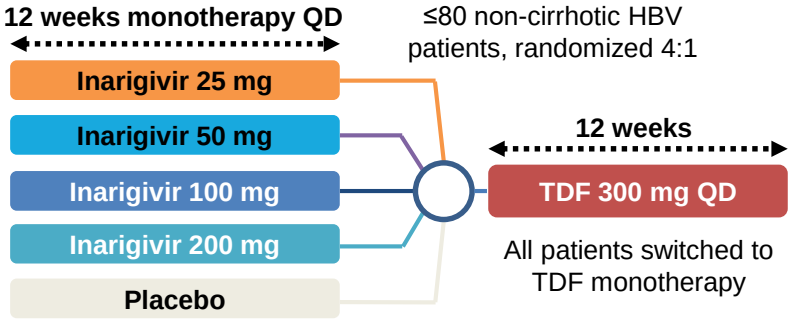
*Combination of RO7049389 and RO7020531 reduced HBsAg level to below LLOQ at the end of treatment in 5 of 7 animals, reduced HBV DNA level to below LLOQ in all animals, which sustained in 4 of 7 during 6-week off-treatment follow-up.

Dai L, et al. ILC 2018, #PS-028

Inarigivir (SB 9200) demonstrates potent dose dependent antiviral activity in HBV naive patients: Role of HBeAg status and BL HBsAg in antiviral response

INARIGIVIR: a RIG–I agonist with a dual mechanism

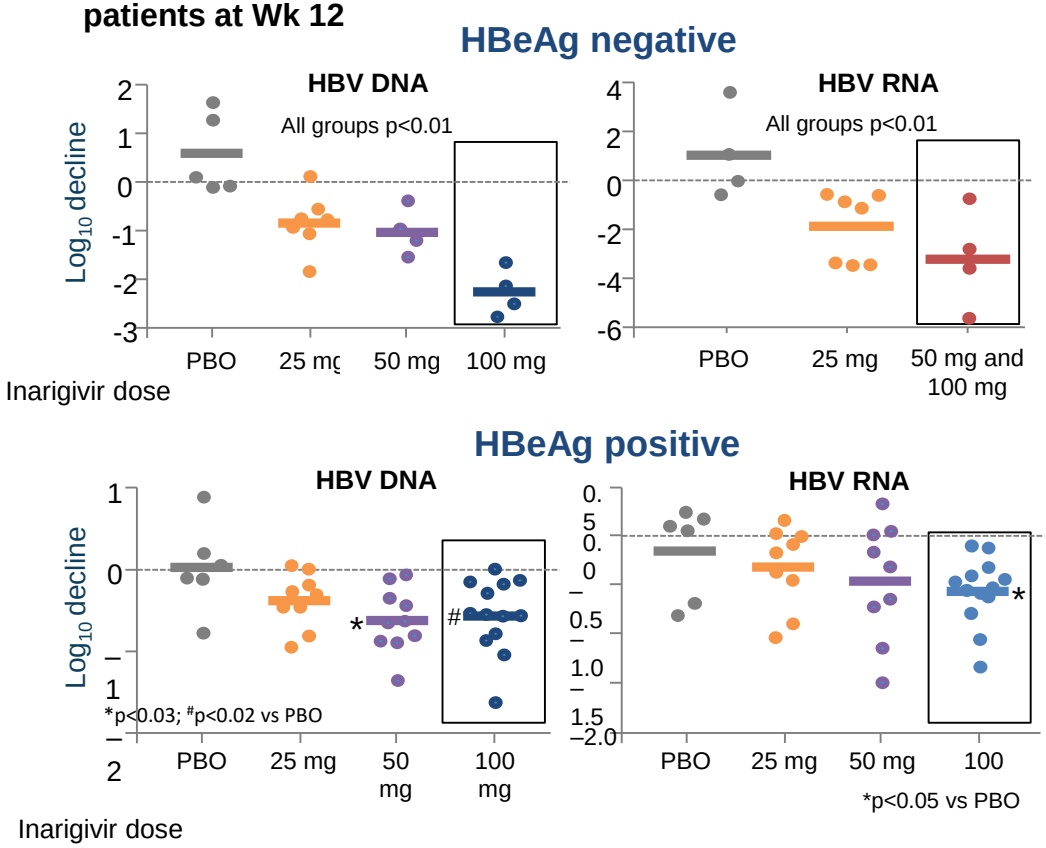
- Restores hepatic selective innate and adaptive immune response stimulating the production of type I and III IFNs
- Inhibits HBV replication complex via direct acting antiviral effect



60 CHB patients:

- 11 pts PBO
- 33 HBeAg pos
- 16 HBeAg neg

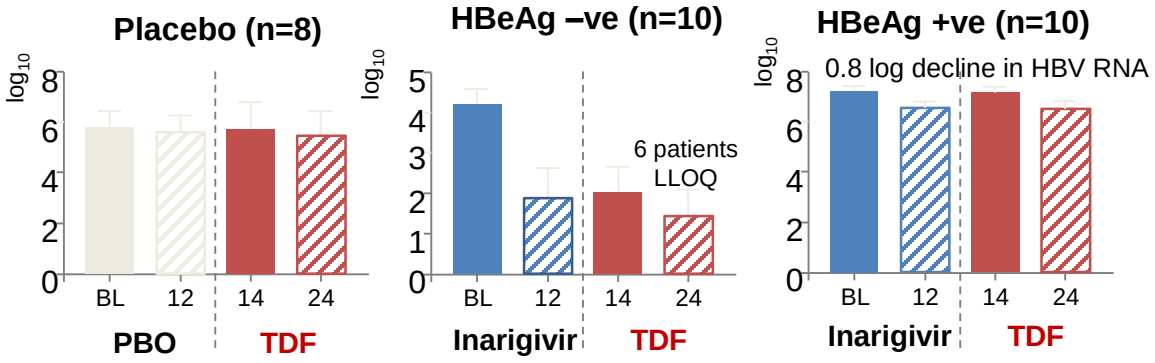
Dose response in both HBeAg negative and HBeAg positive patients at Wk 12

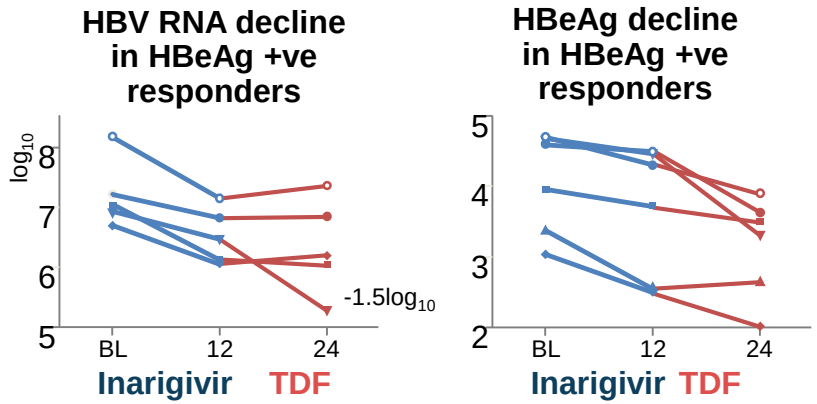


Yuen MF, et al. AASLD 2018, San Francisco, USA. #75

Inarigivir (SB 9200) demonstrates potent dose dependent antiviral activity in HBV naïve patients: Role of HBeAg status and BL HBsAg in antiviral response

HBV RNA reduction after switching to TDF in patients on inarigivir but not PBO



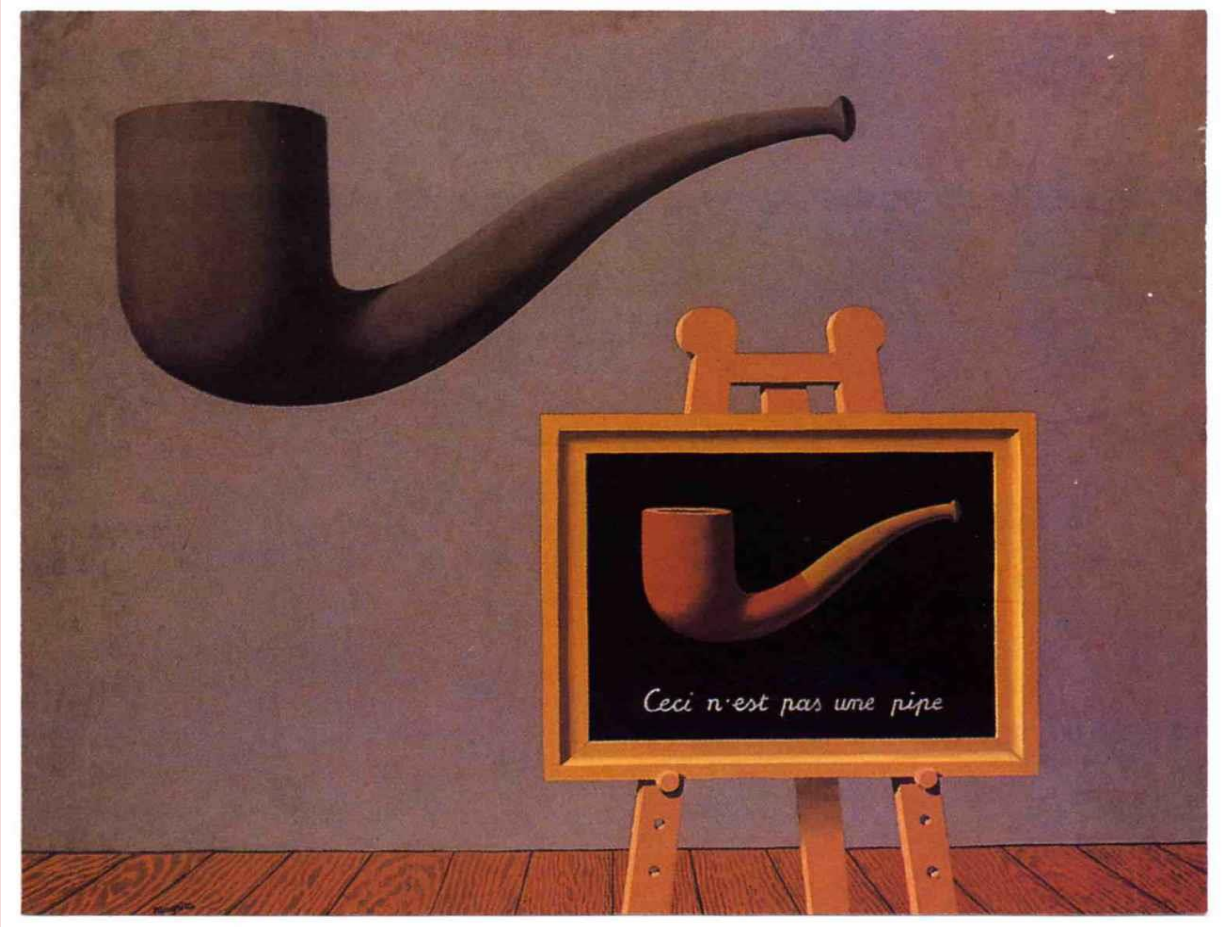


- HBsAg reduction of 0.8 log_{10} (range 0.5–1.4) in 13 responder pts (>0.5 log reduction in HBsAg)
- HBsAg response associated with declines in HBV DNA and HBV RNA
- BL HBsAg <4 logs, BL IP-10 and Wk 12 IP-10 decline predicted HBV DNA and HBV RNA responses
- ALT flares >200 IU/mL in 6 pts on inarigivir (associated with HBV DNA and RNA reduction in 4 wks) and 3 pts on PBO; no changes in bilirubin, INR or with flares

- ➢ Dose-response in HBV RNA and HBsAg reduction with inarigivir
- ➢ Effect more prominent in HBeAg –ve than HBeAg +ve pts
- ➢ ALT flare may indicate immune clearance induced by inarigivir

Unmet need for a personalized management of current and future antiviral treatments

- Identification of the immunologial profile associated with an effective control of HBV infection
- Availability of biomarkers with high diagnostic accuracy in the identification of the carriers who achieved an effective and persistent control of HBV infection



Magritte, I due misteri 1966

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INNOVATIVE MODEL OF
IMPC
PERSONALIZED CARE

Ferruccio Bonino
UPMC Institute for Health
Chianciano