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HBV cure Task Force

ICE-HBV International Coalition to Eliminate HBV

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"Intrahepatic decay of HDV during antiviral therapy"

Miroslava SUBIC-LEVRERO

[1] Lyon Hepatology Institute (IHU Lyon), Lyon, France
[2] Department of Hepatology, Hôpital Croix-Rousse, Hospices Civils de Lyon, Lyon, France

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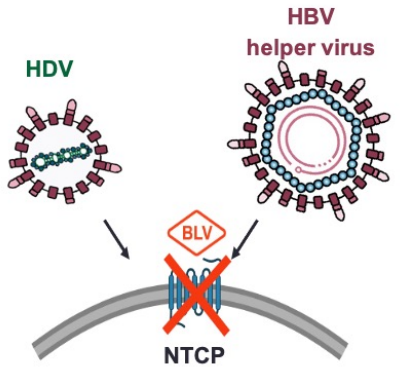
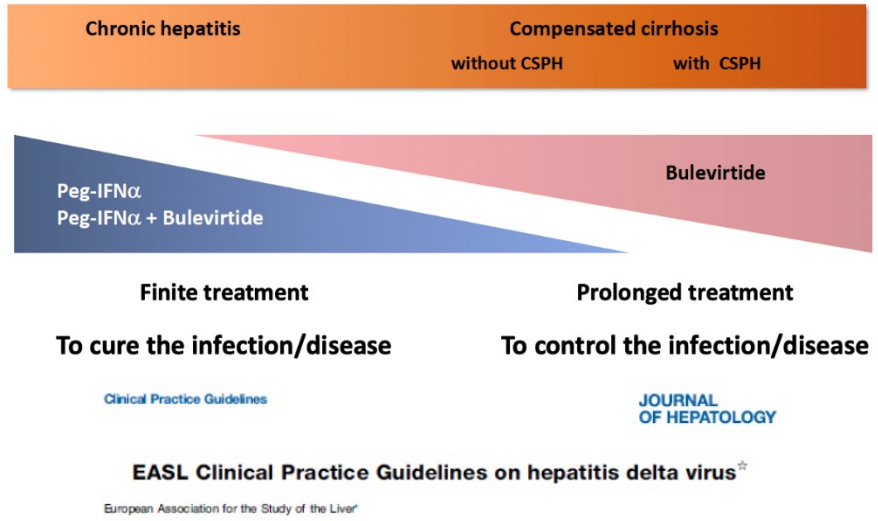
Background: Chronic hepatitis D

- Between 12-20 million people are chronically infected with the hepatitis delta virus (HDV) worldwide[1]
- HDV causes the most severe form of chronic viral hepatitis[2,3]

- HDV requires the envelope protein from hepatitis B virus (HBV) to infect hepatocytes[4]
- Treatment options for chronic hepatitis delta (CHD) are still limited:
  - Pegylated interferon-alfa (PegIFNα) is recommended as off-label therapy[5]
  - The HBV and HDV entry inhibitor bulevirtide (BLV) 2 mg has received full approval in July 2023 in Europe for the treatment of adults with CHD and compensated liver disease
  - PegIFNα + BLV combo explored in clinical trials and real life

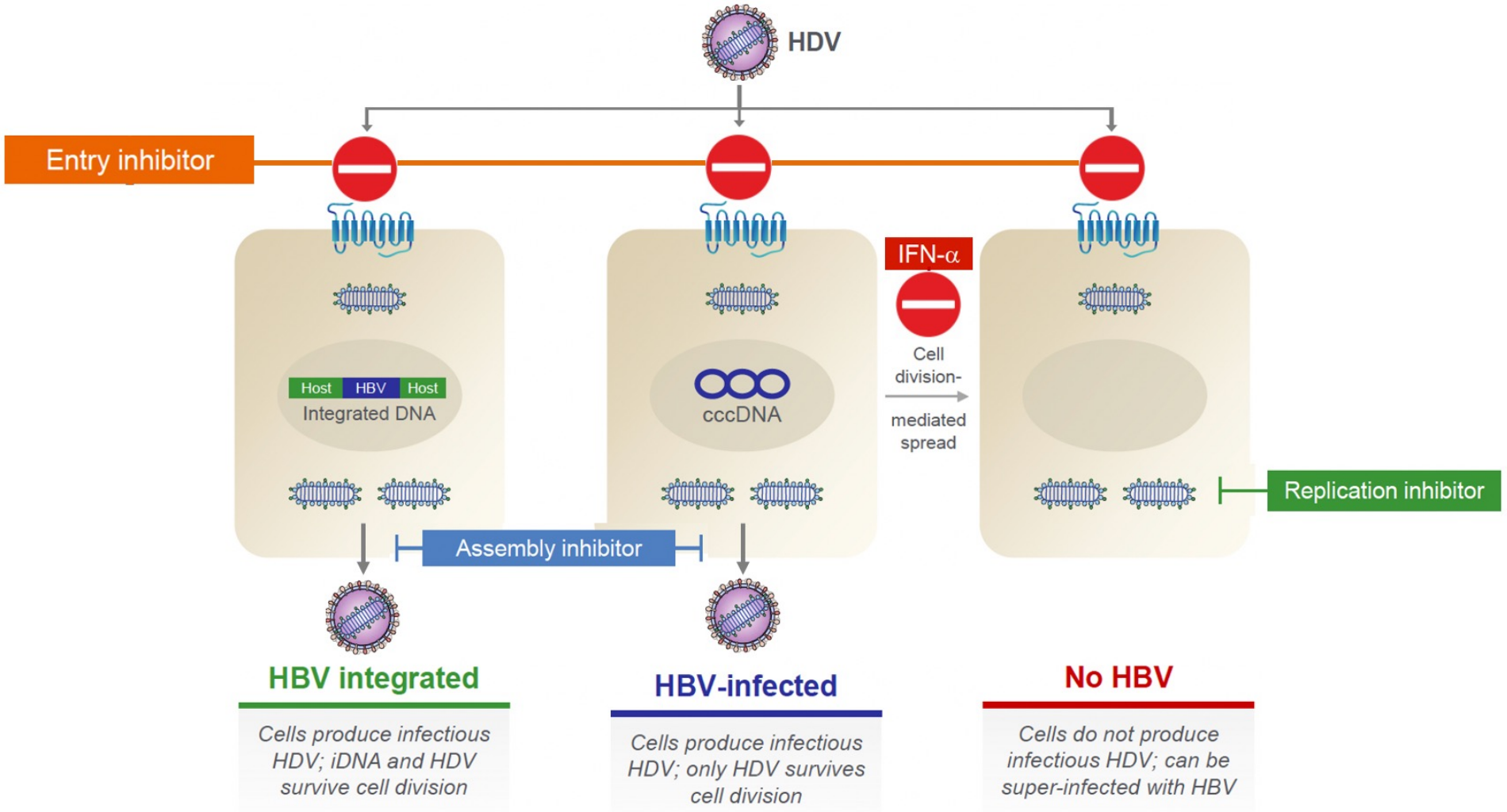




1. Stockdale AJ, et al. J Hepatol 2020;
2. Alfaiate D, et al. J Hepatol. 2020;
3. Rizzetto M, et al. J Hepatol 2021;
4. Asselah T, Rizzetto M. N Eng J Med 2023;
5. Sandmann L, et al. Liver International 2022;00:1-11. 2

modified from Allweiss et al. EASL 2024

Which cells does HDV infects ? How to target all HDV positive cells ?



IFN-α added value in CHD : inhibition of cell-division mediated spread

modified from S Fletcher DZIF Workshop 2024

AGENDA

- ✓ Intrahepatic analysis of BLV monotherapy
  - MYR202, MYR203, MYR301 — Allweiss et al. J Hepatol 2024
  - the « Milan patient » (real life long term treatment) — Anolli et al. J Hep Reports 2023

- ✓ Intrahepatic analysis of BLV – IFN combo — Allweiss et al. EASL 2024
  - MYR204

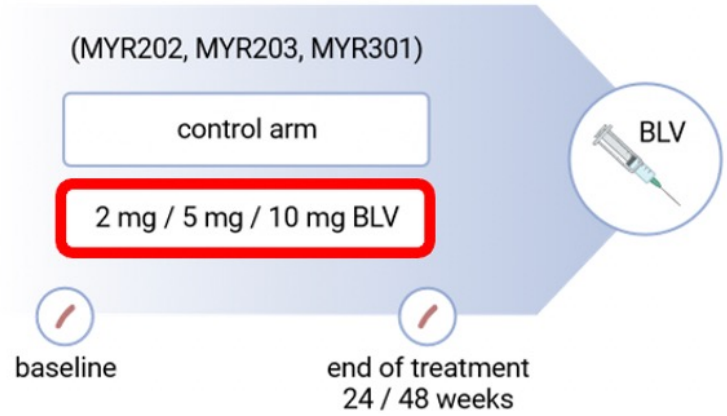
Intrahepatic analyses in BLV trials

Design

BLV monotherapy

Allweiss et al. J Hepatol 2024

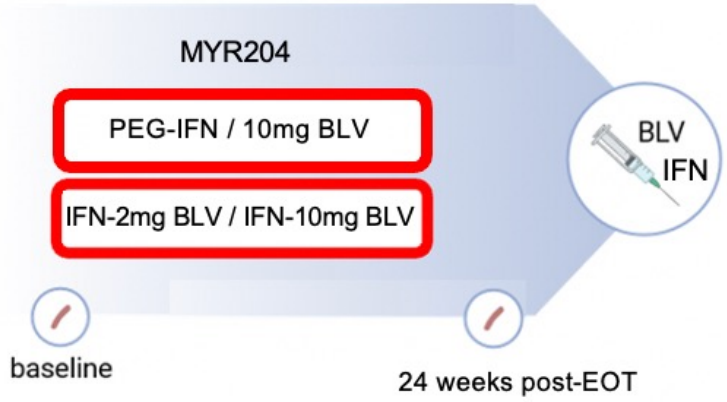
Combined analysis of
126 paired biopsies



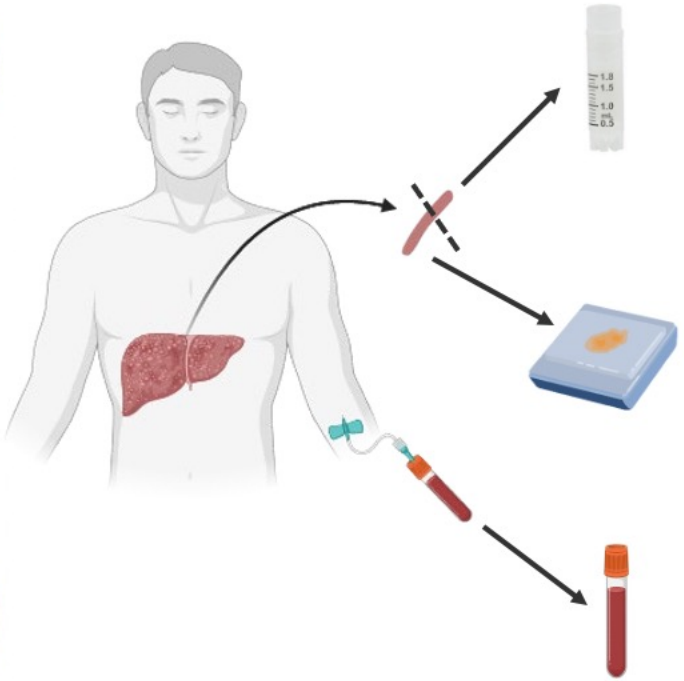
BLV + PEG-IFN

Asselah et al. AASLD 2023
Asselah et al. EASL 2024

Combined analysis of
51 paired biopsies



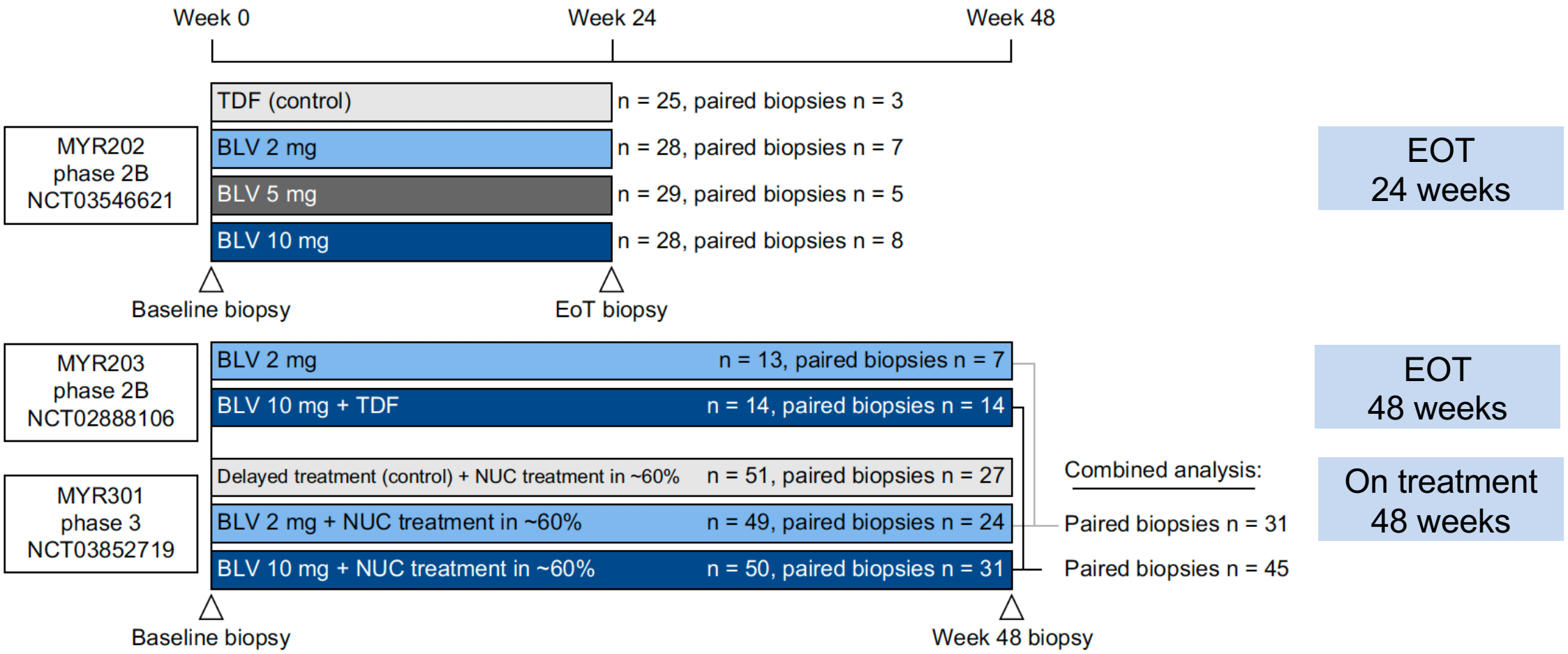
Methods



Frozen tissue stored in Allprotect	qPCR for HDV RNA, HBV RNA and DNA & host gene expression
Formalin-fixed, paraffin-embedded (FFPE) blocks	immunohistochemistry for HDV antigen (HDAg) & quantification of HDAg-positive cells
Peripheral blood	biochemical and virological parameters & Spearman correlation analysis for peripheral (HDV RNA, ALT) vs. intrahepatic parameters (HDV RNA, CXCL10)

modified from Allweiss et al. EASL 2024

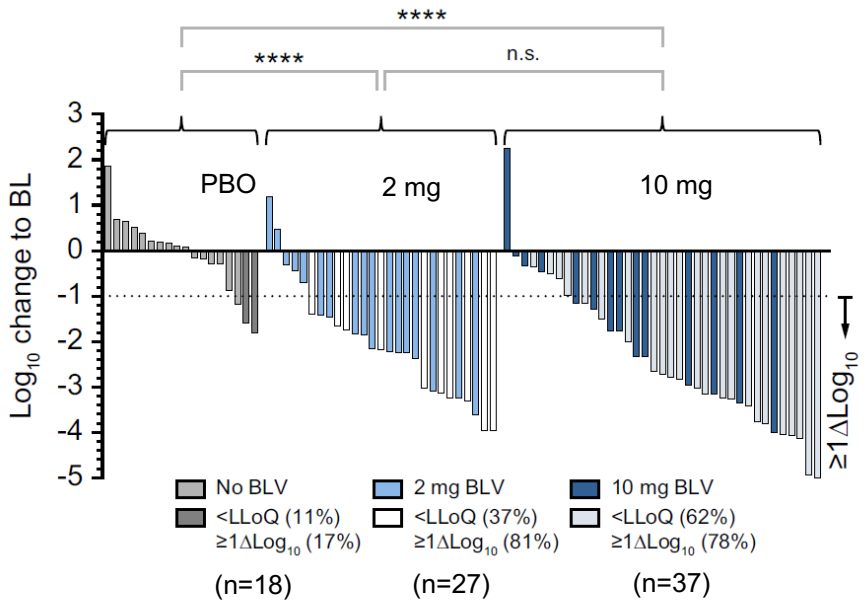
BLV monotherapy studies



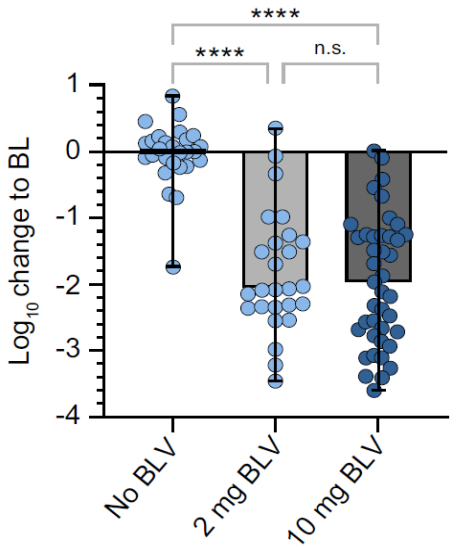
Allweiss et al. J Hepatol 2024

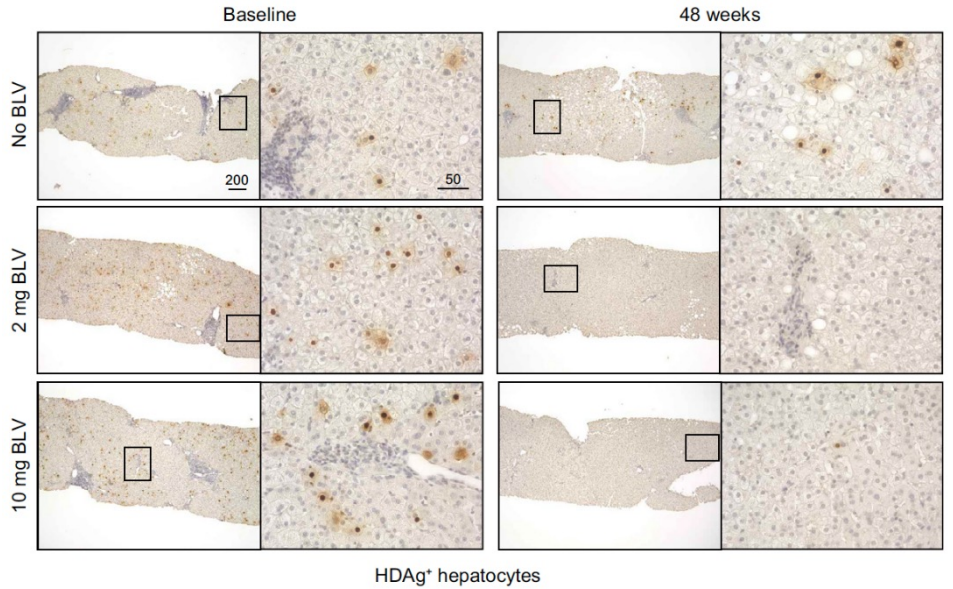
MYR203 and 301 studies
Intrahepatic HDV responses after 48 weeks of BLV monotherapy

Intrahepatic HDV RNA levels



Intrahepatic HDAg levels





- **Serum and liver HDV RNA** levels strongly correlated (Spearman r: 0,62, p <−0.0001)
- **Liver HDV RNA negative**: 11% in control, 37% and 62% in 2 and 10 mg arms, respectively
- **Liver HDAg negative**: 0% in control, 50% and 54% in 2 and 10 mg arms, respectively

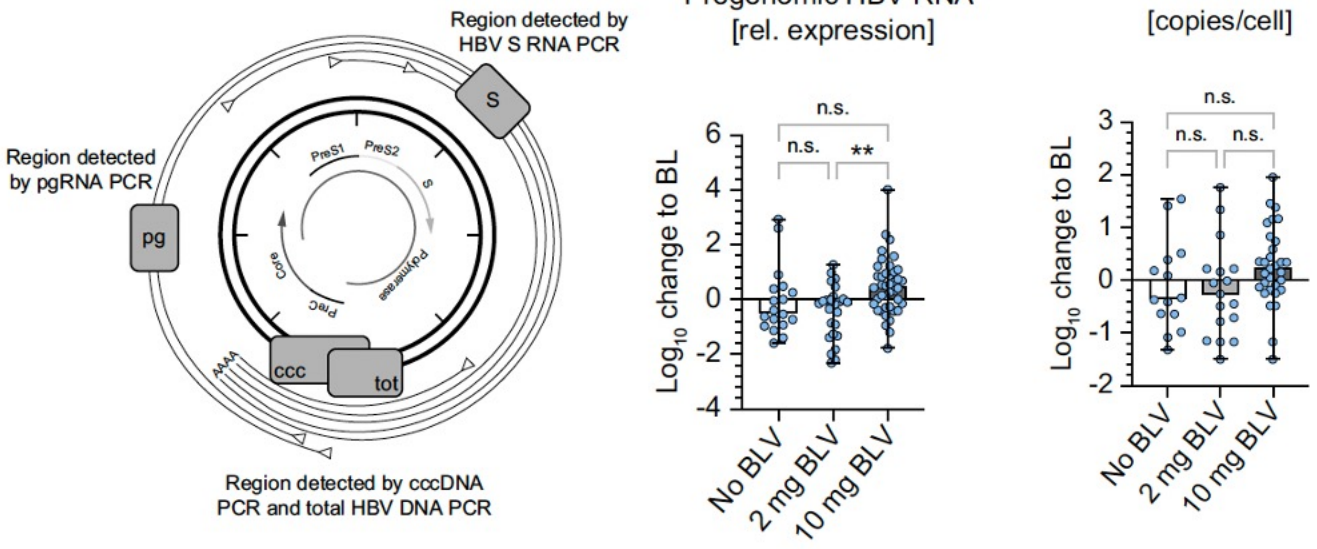
✓ **Blocking viral entry diminishes liver inflammation and promotes a strong reduction of HDV infection within the liver**

✓ **Serum HDV RNA levels are strongly correlated with liver HDV RNA**

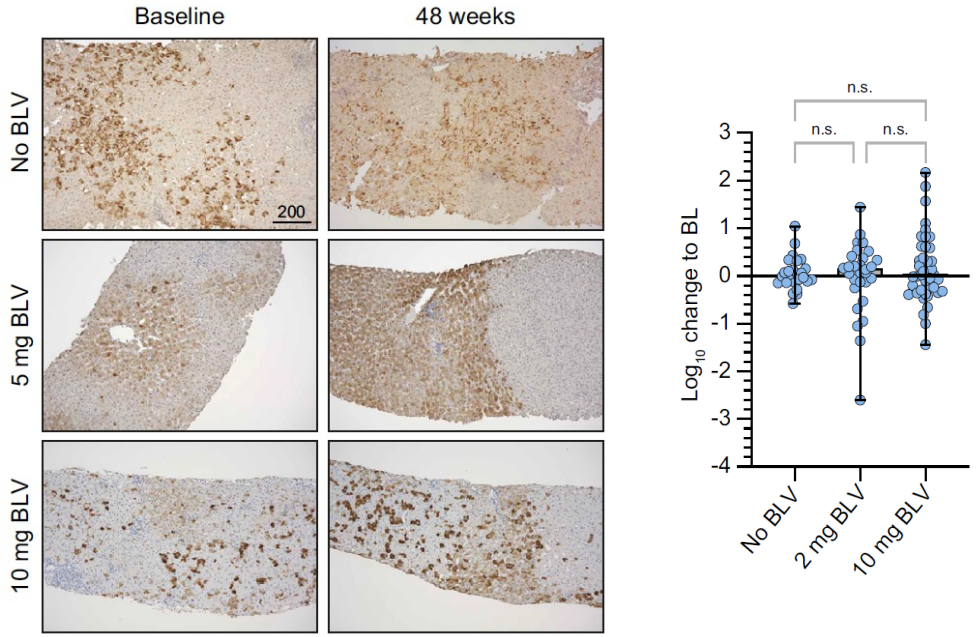
Allweiss et al. J Hepatol 2024

MYR203 and 301 studies
Intrahepatic HBV responses after 48 weeks of BLV monotherapy

Intrahepatic HBV RNA and DNA



Intrahepatic HBsAg levels

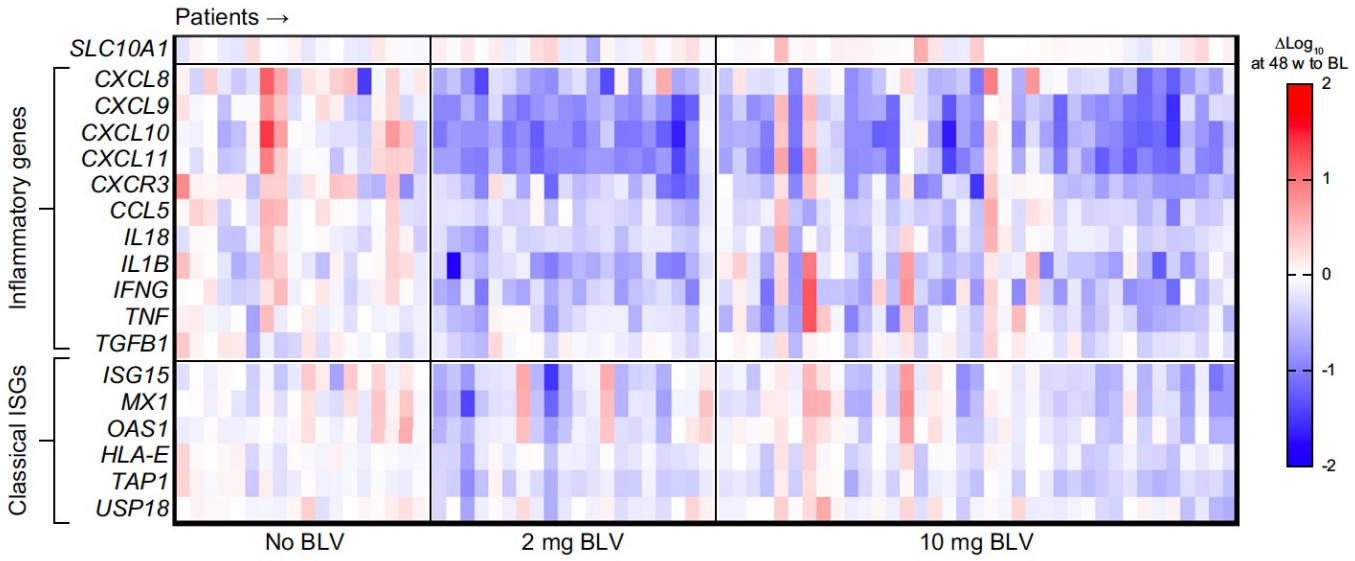


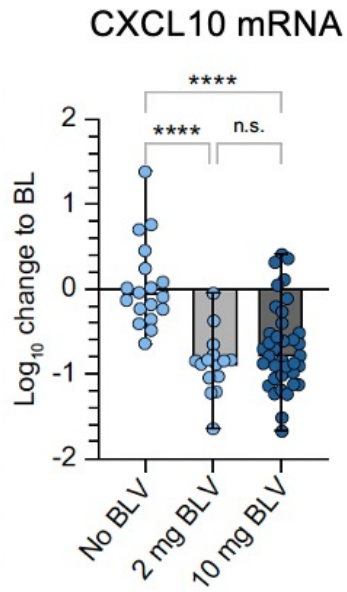
- ✓ No changes in the liver of pgRNA or cccDNA
- ✓ No changes in intrahepatic HBsAg levels

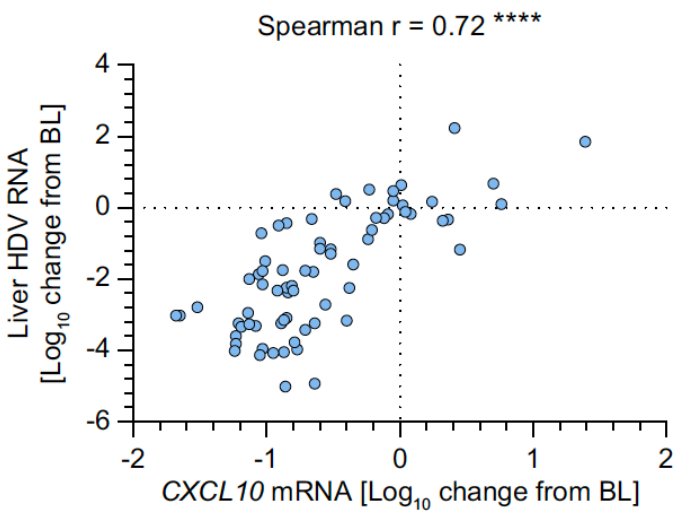
Allweiss et al. J Hepatol 2024

MYR203 and 301 studies
Intrahepatic host gene expression

Intrahepatic expression of inflammatory chemochines and cytokines







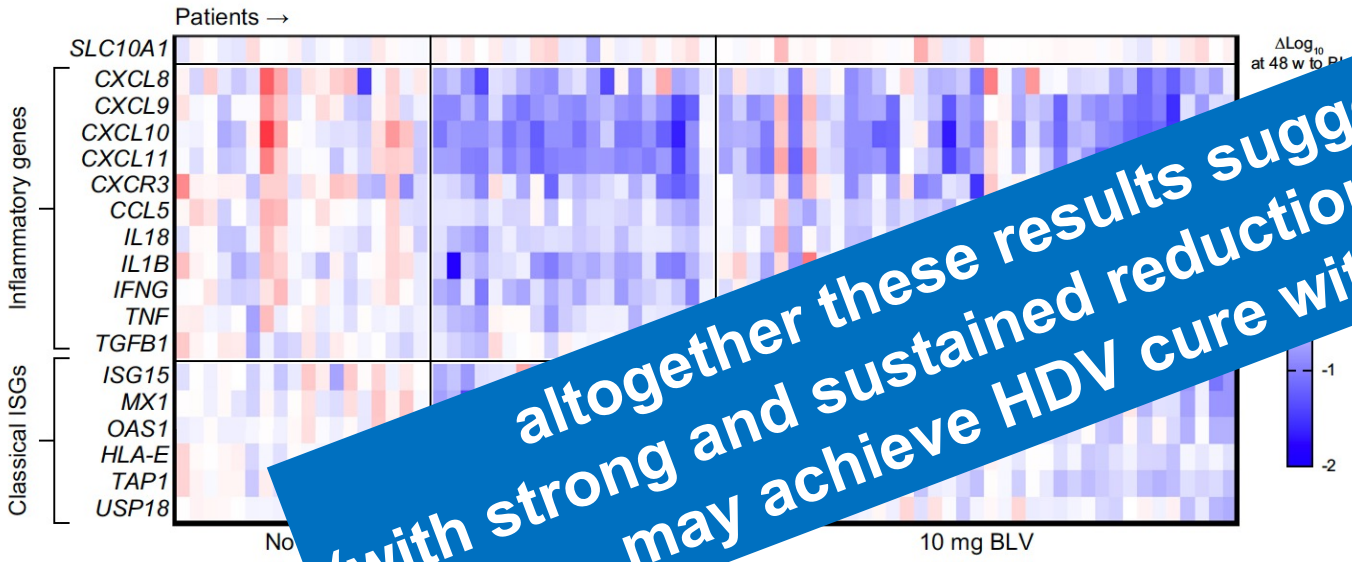
✓ **Expression levels of inflammatory chemokines and cytokines were low after 48 weeks of BLV monotherapy**

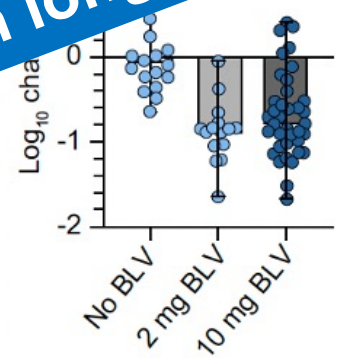
✓ **The reduction of expression levels (CXCL10 mRNA) correlates with the reduction of intrahepatic HDV RNA (and ALT levels).**

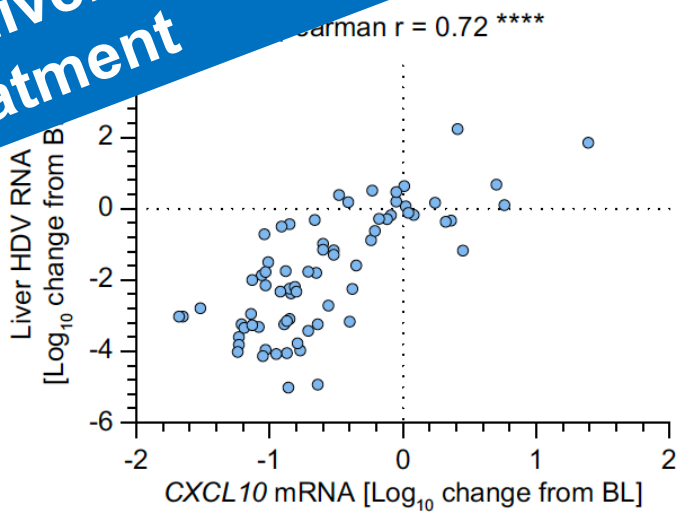
Allweiss et al. J Hepatol 2024

MYR203 and 301 studies
Intrahepatic host gene expression

Intrahepatic expression of inflammatory chemochines and cytokines







altogether these results suggest that some patients (with strong and sustained reduction of serum and liver HDV RNA) may achieve HDV cure with long-term treatment

✓ **Expression l s of inflammatory chemokines and cytokines were low after after 48 weeks of BLV monotherapy**

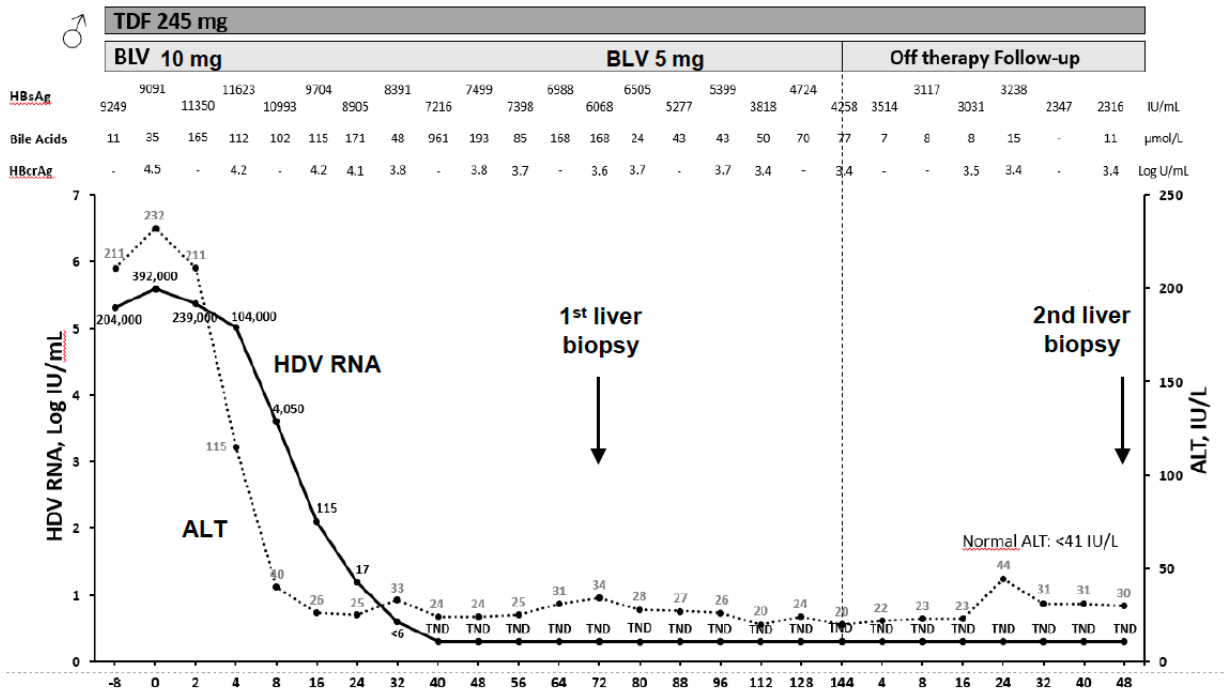
✓ **The reduction of expression levels (CXCL10 mRNA) correlates with the reduction of intrahepatic HDV RNA and ALT levels.**

Allweiss et al. J Hepatol 2024

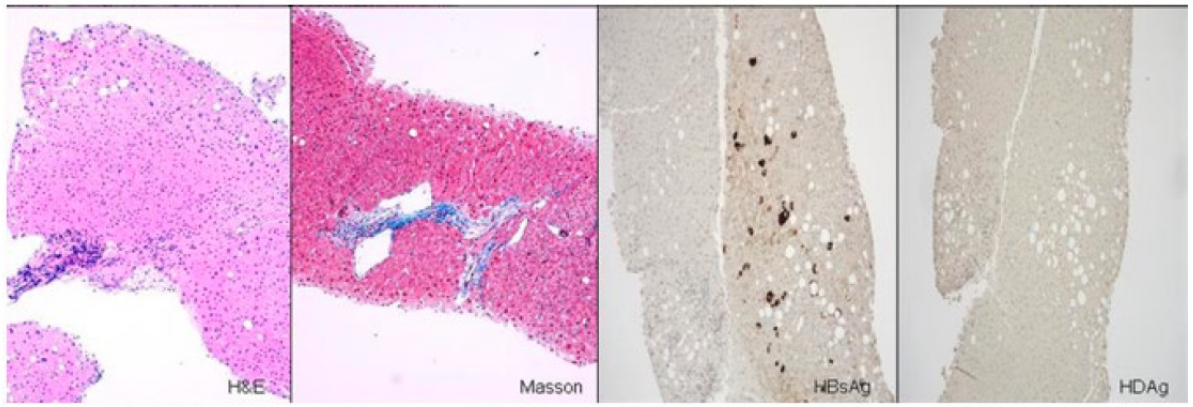
The “Milan patient”
A 3-year course of BLV monotherapy may cure HDV without HBsAg loss

A 55 year-old patient with HDV-related compensated cirrhosis with F1 esophageal varices and contraindications to pegIFNα

Virological and biochemical response during and off BLV therapy



2nd liver biopsy performed at week 48 off-therapy



H&E staining | Masson staining | HBsAg staining | HDAg staining

- Minimal features of inflammation, improvement of fibrosis (Ishak G1 S4) and resolution of autoimmunity features compared to baseline biopsy (Ishak G9 S6)
- HBsAg staining positive (<1%), HBcAg negative.
- **HDAg, HDV RNA and cccDNA undetectable (Dandri’s lab)**
- **HDAg and intrahepatic HDV RNA were already undetectable** in the liver biopsy performed on-therapy at week 72 (Dandri’s lab)

Clinical outcomes

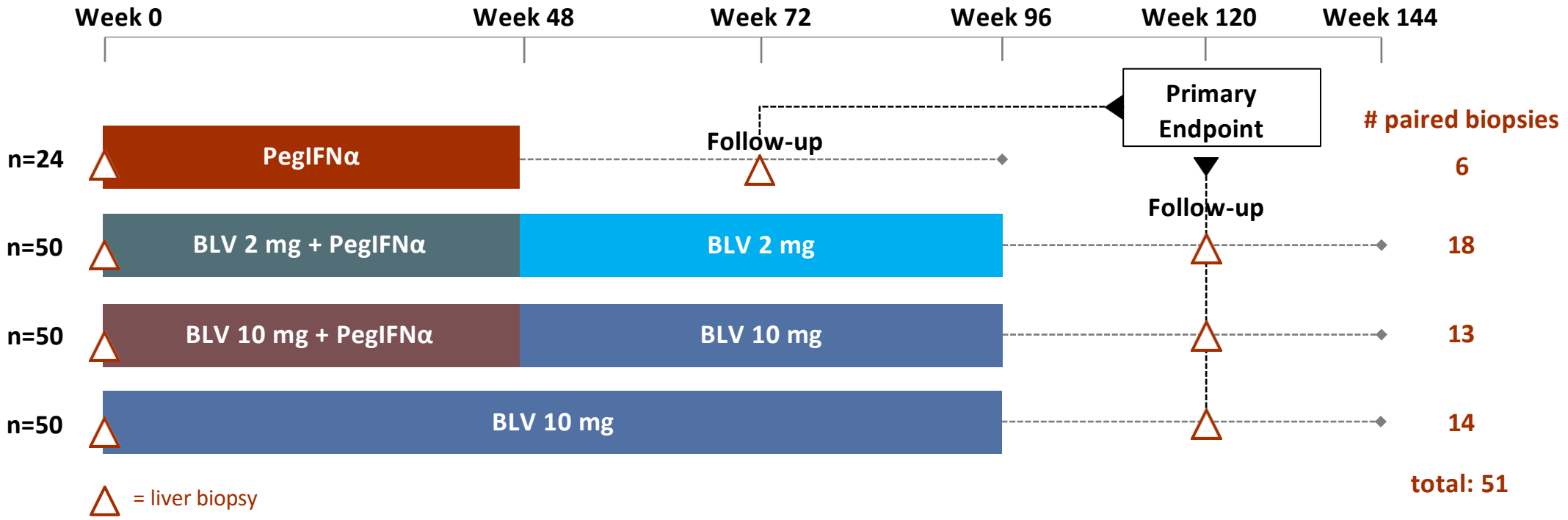
- HDV suppression/cure resulted in a significant improvement in biochemistry, liver function parameters, AFP, LSM, and in regression of esophageal varices.
- No specific safety issues, BA normalized after BLV discontinuation

Conclusions

- **A 3-year course of BLV monotherapy may cure HDV infection even in difficult-to-treat patients with advanced compensated cirrhosis**
- **HDV eradication occurred without HBsAg loss**

Anolli MP et al. J Hepatol 2023

MYR204 study
Intrahepatic responses 24 weeks after EOT (PEG-IFN + BLV)

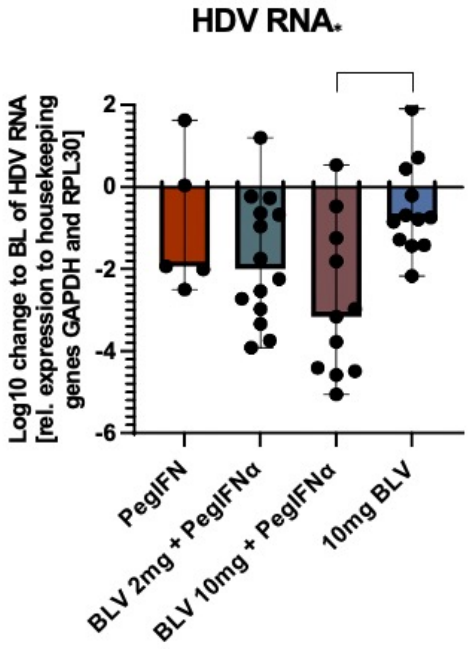


Open-label, randomized, multicenter, phase 2b study (NCT03852433) conducted in 19 sites across 4 countries (France, Moldova, Romania, and Russia)

Asselah et al. NEJM 2024
Allweiss et al. EASL 2024

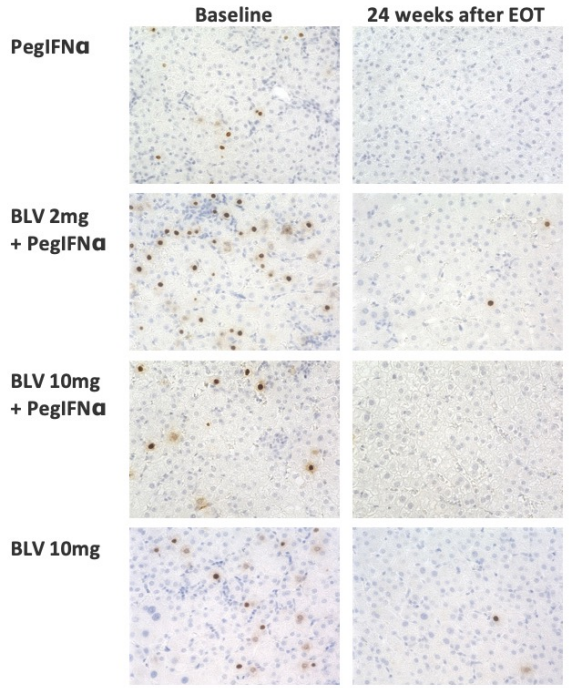
MYR204
Intrahepatic HDV responses 24 weeks after EOT

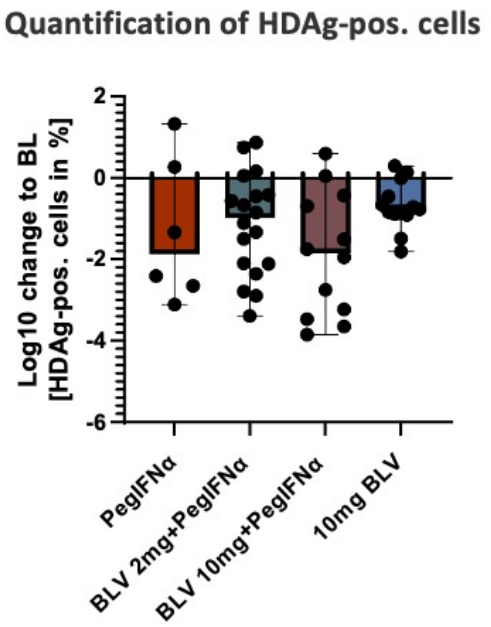
Intrahepatic HDV RNA levels



- Kruskall-Wallis test with Dunn as a post-test for differences of Log10 change from baseline between arms (bar graphs);
- LLoQ 0.0001 relative expression
- p values ≤0.05 depicted as *; p values ≤0.01 as **
  Caveat: Study was not powered for this analysis

Intrahepatic HDAg levels

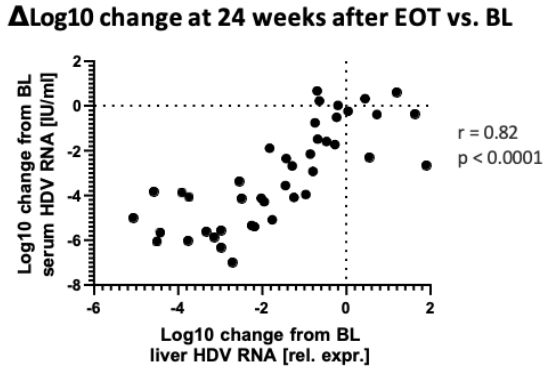




- Wilcoxon matched pairs test for differences between baseline and follow-up biopsies
- p values ≤0.05 depicted as *;
- Caveat: Study was not powered for this analysis; LLoQ 0.012% HDAg-positive cells

Correlation serum vs liver

HDV RNA in serum vs. liver



Serum HDV RNA levels correlate with liver HDV RNA levels

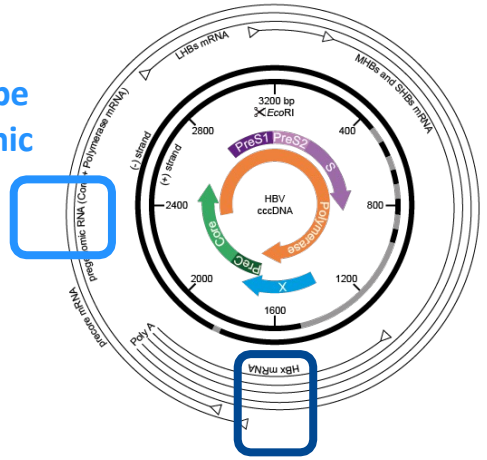
- ✓ **HDV RNA levels and HDAg-positive cells in the liver were reduced at 24w after EOT**
- ✓ **Intrahepatic HDV RNA levels strongly correlated with the number of HDAg positive cells suggesting that BLV treatment reduced the number of infected cells**

The highest HDV RNA off-treatment decline in the BLV 10mg + PegIFNα combination arm

Allweiss et al. EASL 2024

MYR204
Intrahepatic HBV responses 24 weeks after EOT

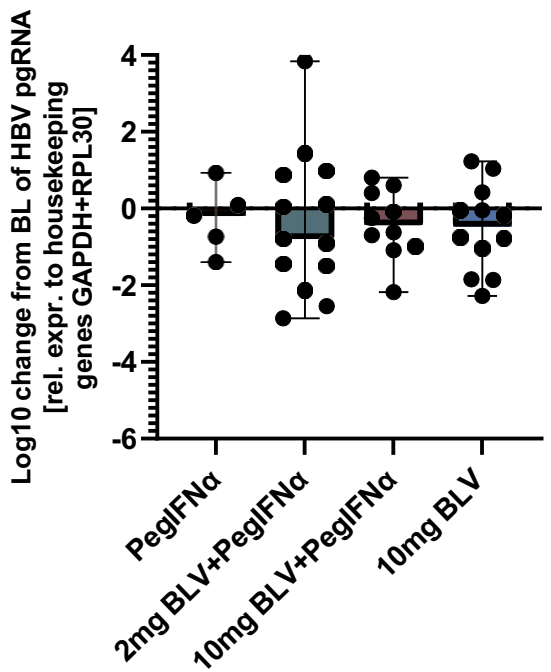
primer/probe in pregenomic RNA



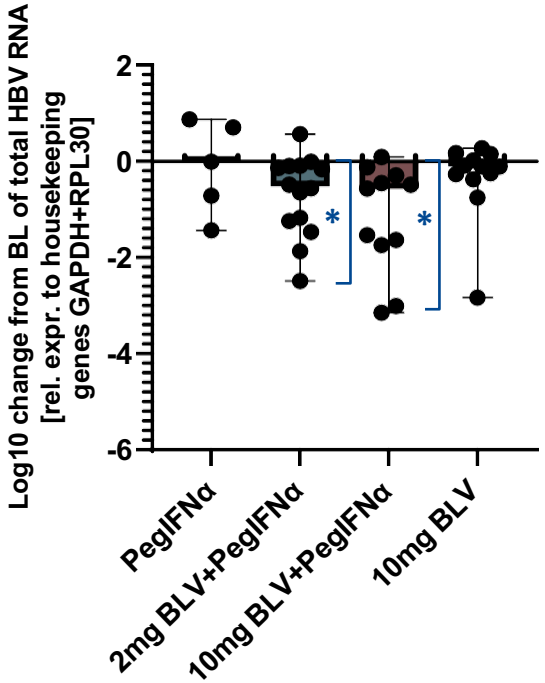
primer/probe in common region for all HBV RNAs

- ✓ Wilcoxon matched pairs test for differences between baseline and follow-up biopsies
- ✓ p values ≤0.05 depicted as *
  Caveat: Study was not powered for this analysis.

HBV pregenomic RNA



Total HBV RNA

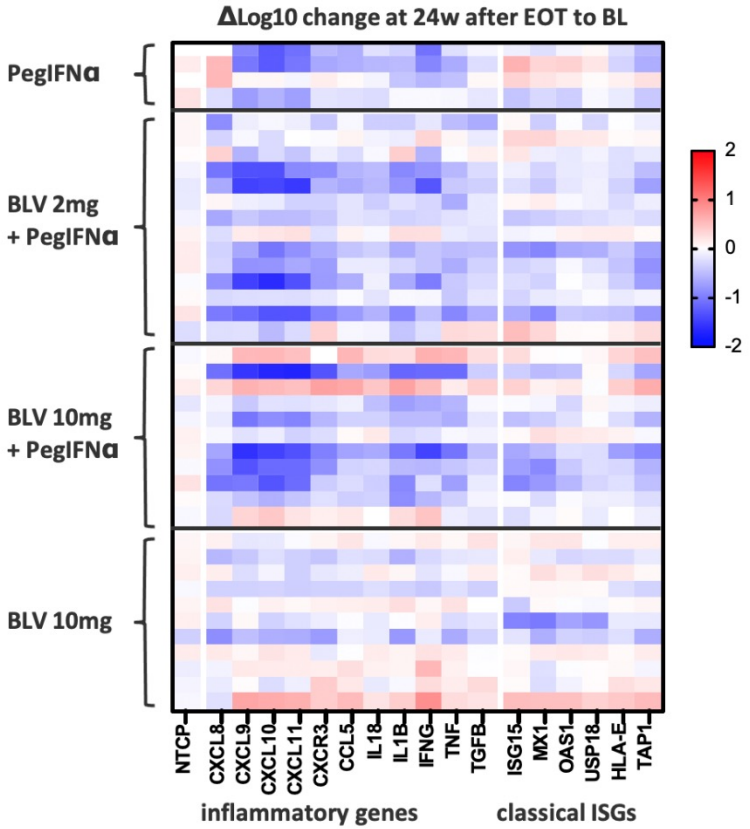


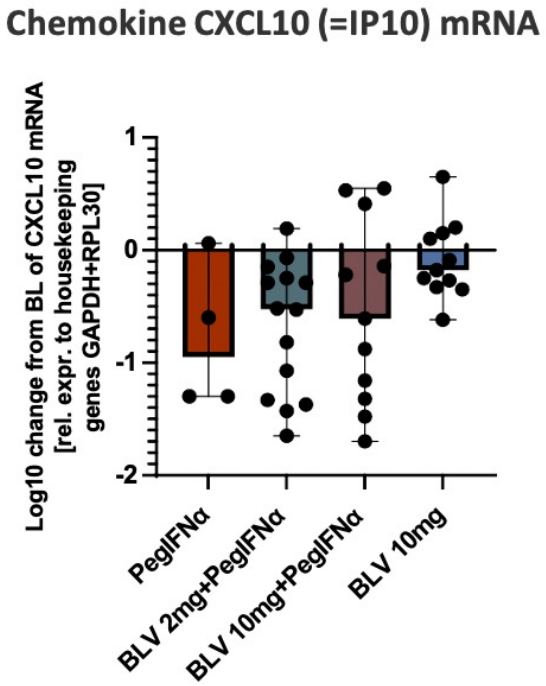
- ✓ **HBV RNA levels did not change significantly between BL and 24 weeks after EOT.**
- ✓ **Nevertheless, there were modest median reductions of total HBV RNA in the combination arms.**

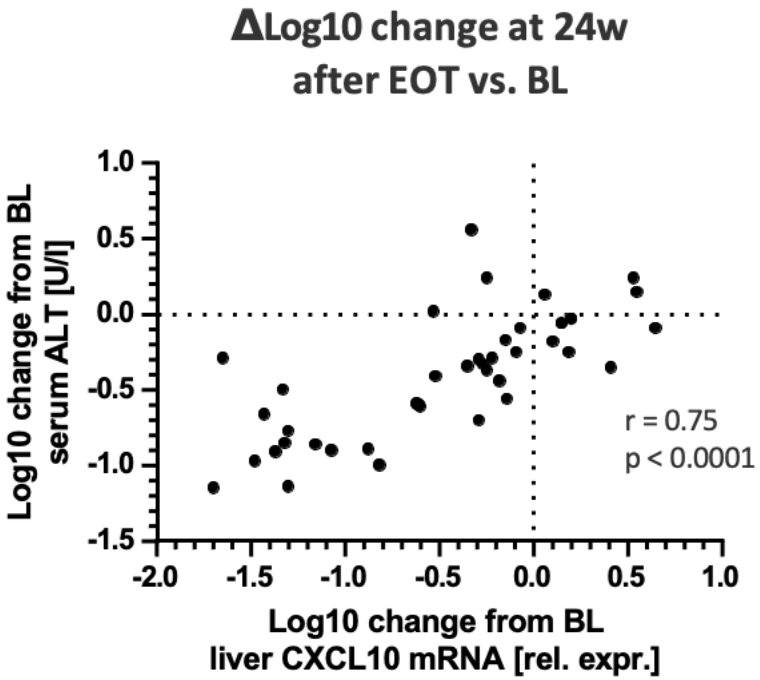
Allweiss et al. EASL 2024

MYR204
Intrahepatic host gene expression

Intrahepatic expression of inflammatory chemokines and cytokines







✓ **Expression levels of inflammatory chemokines and cytokines were low after combination therapy and PegIFN monotherapy at 24 weeks after EOT.**

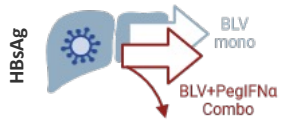
✓ **The reduction of expression levels (CXCL10 mRNA) correlates with the reduction of intrahepatic HDV RNA (and ALT levels).**

Allweiss et al. EASL 2024

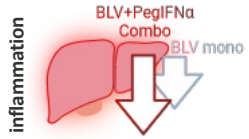
Summary & Conclusions

Intrahepatic analysis in paired BL vs on treatment and post-treatment biopsies demonstrated a strong correlation between intrahepatic and serum HDV RNA reductions both in BLV mono and PEG-IFNα /BLV combo

Similar to serological findings, the highest rate of off-treatment virologic response in the liver was observed in the arm that received combination of BLV 10mg + PegIFNα.



Intrahepatic HBV markers did not show consistent changes, with the exception of few patients in the PegIFNα arms with reduced HBV markers.



Concomitant with the decrease of HDV parameters, expression levels of innate immune genes declined

Limitations:

- ✓ No intrahepatic data yet for the MYR 301 BLV monotherapy off treatment
- ✓ Lack of biopsies at end of treatment in the MYR 204 study do not allow for the analysis of on-treatment efficacy in the liver.
- ✓ Relatively low number of paired biopsies in some treatment arms (e.g., PegIFN monotherapyiarm in MYR 204)

modified from Allweiss et al. EASL 2024