# COMMON HEPATOLOGY CONSULTS (HYPERFERRITINEMA AND HEPATITIS B)

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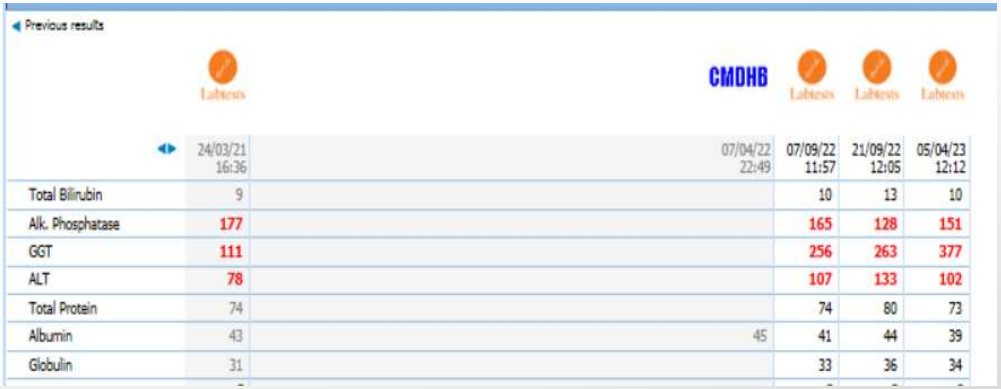
# CASE 1: MR SC

- 57YO MALE
- KNOWN NAFLD WITH STAGE II FIBROSIS ON LIVER BX 2016
- GP REFERRAL: DERANGED LFTS AND GROSSLY RAISED FERRITIN



Previous results

CMDHB

| | 24/03/21 16:36 | 07/04/22 22:49 | 07/09/22 11:57 | 21/09/22 12:05 | 05/04/23 12:12 |
|---|---|---|---|---|---|
| Total Bilirubin | 9 | | 10 | 13 | 10 |
| Alk. Phosphatase | 177 | | 165 | 128 | 151 |
| GGT | 111 | | 256 | 263 | 377 |
| ALT | 78 | | 107 | 133 | 102 |
| Total Protein | 74 | | 74 | 80 | 73 |
| Albumin | 43 | 45 | 41 | 44 | 39 |
| Globulin | 31 | | 33 | 36 | 34 |

CMDHB

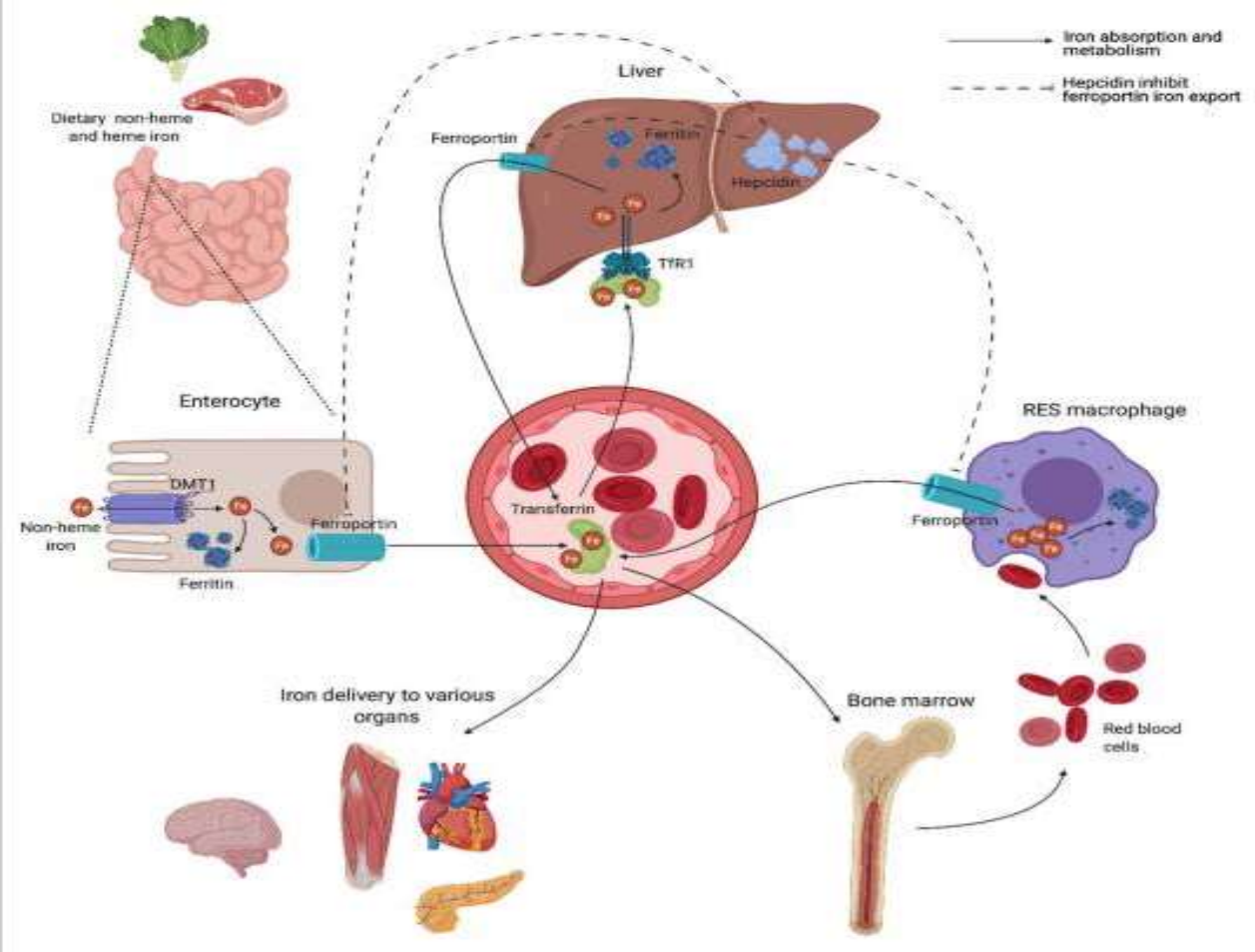
| | 14/12/11 11:42 | 21/03/14 | 26/08/15 12:37 | 25/06/16 09:39 | 23/08/17 11:22 | 09/05/18 14:11 | 23/01/19 13:31 | 19/02/20 14:26 | 14/07/20 02:30 | 05/08/20 13:15 | 24/03/21 16:36 | 07/09/22 11:57 | 21/09/22 12:05 | 05/04/23 12:12 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Serum Iron | 23 | 22 | 29 | 20 | 18 | 15 | 24 | 28 | 21.0 | 15 | 25 | 31 | 31 | 36 |
| Transferrin | 3.3 | 3.2 | 3.0 | 3.2 | 3.1 | 2.9 | 2.6 | 2.9 | | 2.8 | 2.8 | 2.9 | 3.0 | 2.6 |
| Transferrin Saturation | 0.28 | 0.27 | 0.38 | 0.25 | 0.23 | 0.21 | 0.37 | 0.38 | | 0.21 | 0.36 | 0.43 | 0.41 | 0.55 |
| Ferritin | 856 | 1722 | 1206 | 1535 | 1050 | 938 | 1226 | 1353 | 3161 | 2160 | 2326 | 2738 | 3819 | 3000 |
| Comment | | | | | | | | | | | | | | |
| Transferrin | | | | | | | | | 2.5 | | | | | |
| Transferrin saturation | | | | | | | | | 0.37 | | | | | |
| Authorised by | | | | | | | | | IT3000 MMH | | | | | |

# CASE 1: WHAT SHOULD YOU DO NOW??

- A) HEMOCHROMATOSIS GENE TESTING
- B) CHECK HBA1C
- C) CHECK ETOH INTAKE
- D) LIVER BIOPSY
- E) VENESECTION

# IRON HEMOSTASIS

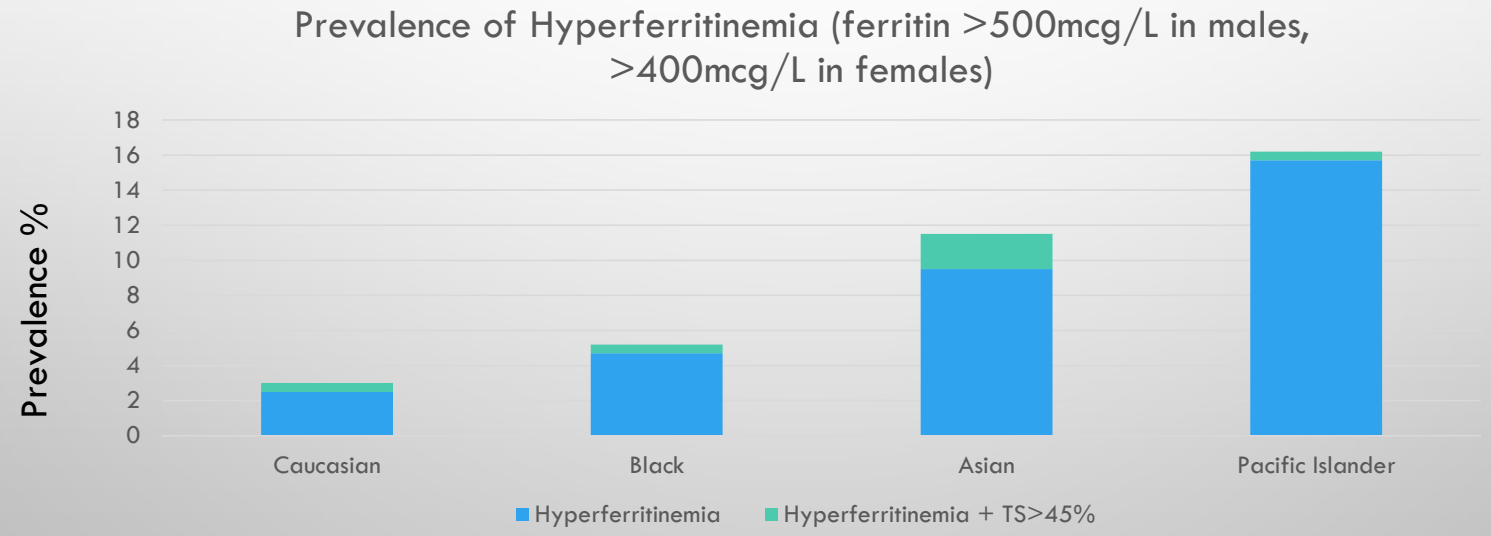
- DIETARY IRON IS ABSORBED AT APICAL SITE OF ENTEROCYTES OF THE PROXIMAL DUODENUM THROUGH DIVALENT METAL TRANSPORTER 1 (DMT1)
- ONE PORTION REMAINS STORED AS FERRITIN INSIDE THE ENTEROCYTE
- REST IS TRANSFERRED THROUGH BASOLATERAL SITE VIA FERROPORRTIN
- IRON BINDS TRANSFERRIN AND IS FURTHER DISTRIBUTED THROUGHOUT THE BODY.
- MOST IRON DISTRIBUTED TO BONE MARROW FOR HB PRODUCTION
- IRON SUBSEQUENTLY RETURNS TO THE CIRCULATION WHEN RBC PHAGOCYTOSED BY MACROPHAGES OF THE RES
- HEPCIDIN PRODUCED IN LIVER REGULATES SYSTEMIC IRON BALANCE THROUGH BINDING OF FERROPORRTIN


Iron absorption and metabolism
Hepcidin inhibit ferroportin iron export
Liver
Ferroportin
Ferritin
Hepcidin
TfR1
Dietary non-heme and heme iron
Enterocyte
DMT1
Non-heme iron
Ferroportin
Ferritin
Transferrin
RES macrophage
Ferroportin
Iron delivery to various organs
Bone marrow
Red blood cells

- IRRESPECTIVE OF LEVELS, IRON IS ELIMINATED AT A BASAL RATE THROUGH DESQUAMATION OF SKIN AND INTESTINAL EPITHELIUM, AND THROUGH BLOOD LOSS IN FERTILE WOMEN
- SYSTEMIC REGULATION OF AVAILABILITY TO MEET IRON NEEDS ARE PREDOMINANTLY MEDIATED THROUGH HEPCIDIN AND THE HEPCIDIN-FERROPORRTIN AXIS

# HOW COMMON IS HYPERFERRITENEMIA?

- HEREDITARY HEMOCHROMATOSIS AND IRON OVERLOAD SCREENING STUDY- HEIRS
- 101,168 MULTI-ETHNIC PRIMARY CARE PARTICIPANTS IN US/CANADA




Adams, NEJM 2005, 352(17):1769-78.

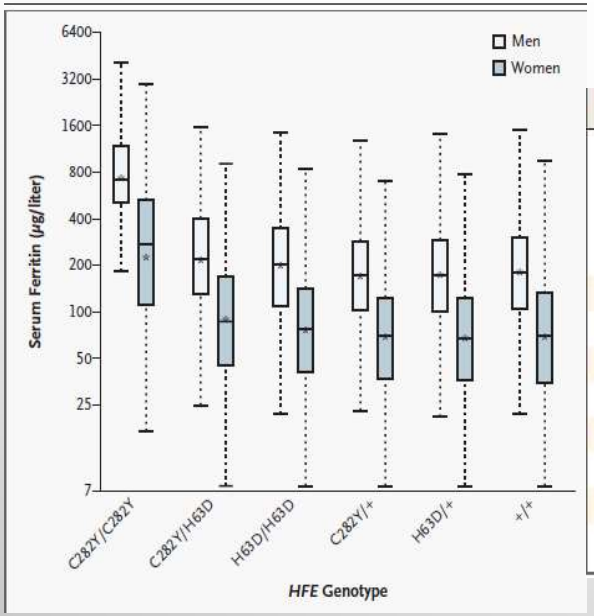



**Table 1. Prevalence of *HFE* C282Y and H63D Genotypes According to Race or Ethnic Group.***

| Race or Ethnic Group | Total No. of Participants | C282Y/C282Y | | C282Y/H63D | | H63D/H63D | |
|---|---|---|---|---|---|---|---|
| | | No. | Prevalence (95% CI) | No. | Prevalence (95% CI) | No. | Prevalence (95% CI) |
| | | | % | | % | | % |
| White | 44,082 | 281 | 0.44 (0.42–0.47) | 908 | 2.0 (2.0–2.1) | 1029 | 2.4 (2.3–2.4) |
| Native American | 648 | 1 | 0.11 (0.061–0.20) | 7 | 0.77 (0.56–1.1) | 7 | 1.3 (0.98–1.8) |
| Hispanic | 12,459 | 7 | 0.027 (0.022–0.032) | 48 | 0.33 (0.30–0.37) | 154 | 1.1 (0.98–1.1) |
| Black | 27,124 | 4 | 0.014 (0.012–0.017) | 35 | 0.071 (0.065–0.078) | 30 | 0.089 (0.081–0.097) |
| Pacific Islander | 698 | 0 | 0.012 (0.0043–0.032) | 0 | 0.096 (0.055–0.17) | 0 | 0.20 (0.12–0.32) |
| Asian | 12,772 | 0 | 0.000039 (0.000015–0.00010) | 0 | 0.0055 (0.0029–0.0093) | 29 | 0.20 (0.17–0.22) |
| Multiple/unknown | 1928 | 6 | — | 19 | — | 21 | — |
| All | 99,711 | 299 | — | 1017 | — | 1270 | — |

Serum ferritin levels in men and women according to genotype

# WHAT ARE THE DRIVERS OF HYPERFERRITINEMIA?

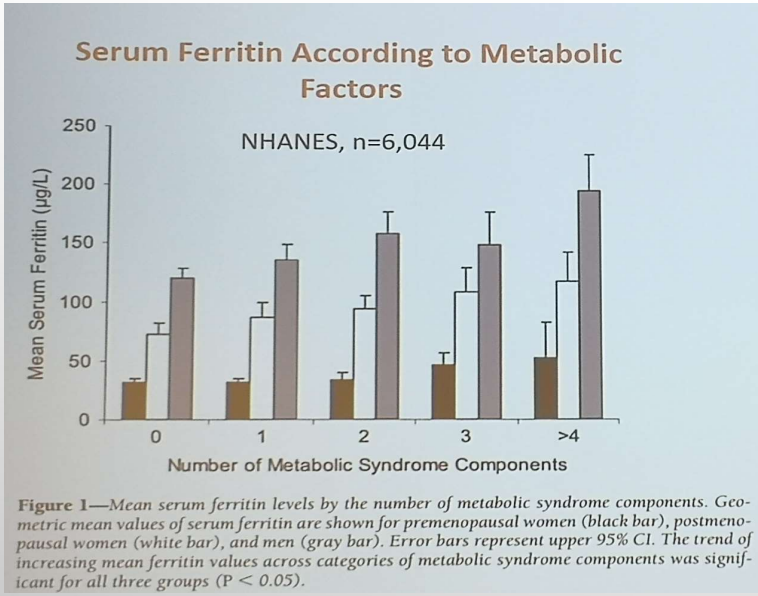
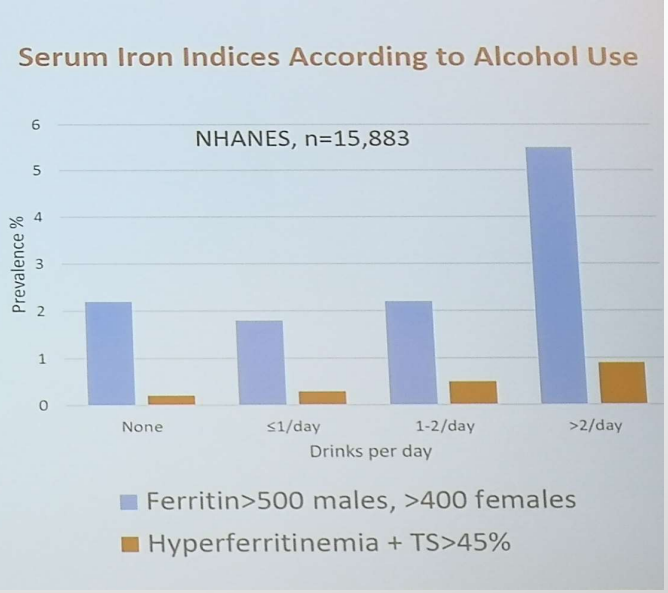
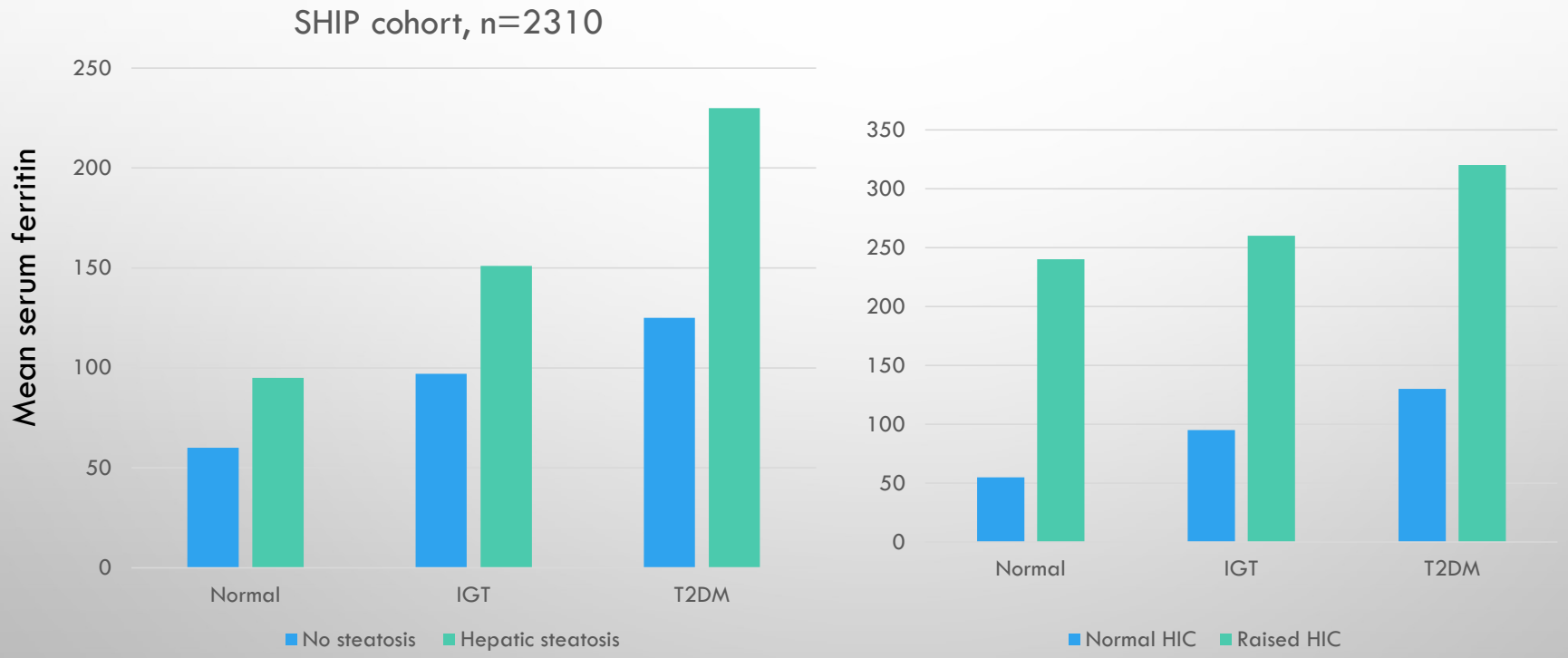



**Figure 1**—*Mean serum ferritin levels by the number of metabolic syndrome components. Geometric mean values of serum ferritin are shown for premenopausal women (black bar), postmenopausal women (white bar), and men (gray bar). Error bars represent upper 95% CI. The trend of increasing mean ferritin values across categories of metabolic syndrome components was significant for all three groups ($P < 0.05$).*




Jehn M et al, Diabetes care, 2007; Ioannou et al, Gastro 2004

# DRIVERS OF HYPERFERRITINEMIA: HEPATIC STEATOSIS, HEPATIC IRON, HYPERGLYCEMIA




Pitchika et al. Liver International Care 2021.

Raised hepatic iron content = R2*>41 sec/-1

# METABOLIC HYPERFERRITINEMIA/ DYSMETABOLIC IRON OVERLOAD SYNDROME

**DIAGNOSTIC CRITERIA**

ELEVATED FERRITIN

FATTY LIVER OR DIABETES OR OVERWEIGHT/OBESITY OR TWO METABOLIC SYNDROME FEATURES

**EXCLUSION CRITERIA**

GENETIC HEMOCHROMATOSIS OR 2* IRON OVERLOAD (TS>50%)

EXCESS ALCOHOL (>60GM/DAY MALES, >40GM/DAY FEFMALES)

ESRF

Valenti et al, Nat Rev Endocrin.

# RISK FACTORS

- MALE
- AGE
- MODERATE ALCOHOL
- GENETIC VARIANTS (PCSK7, SERPINA1, BETAGLOBIN, NMBR, CERULOPLASMIN)

# SPECTRUM OF IRON METABOLISM IN INDIVIDUALS WITH METABOLIC DYSFUNCTION

| Normal iron metabolism | | Metabolic Hyper ferritinemia | Dysmetabolic iron accumulation | Dysmetabolic iron overload syndrome |
|---|---|---|---|---|
| Ferritin (ng/mL) | 50-ULN | ULN-550 | 550-1000 | >1000 |
| Hepatic iron stores (R2* 1/s) | <70 | <70 | 70-140 | >140 |
| Iron stores (vs normal) | - | - | + | ++ |
| Organ damage | - | - | - | + |

# NON INVASIVE NAFLD FIBROSIS SCORES

- 1) NAFLD FIBROSIS SCORE: AGE, BMI, HYPERGLYCEMIA, AST/ALT, PLATELET, ALBUMIN
- 2) AST/PLATELET RATIO INDEX : AST, PLATELET
- 3) FIB-4 SCORE: AGE, AST, PLATELET, ALT
- 4) BARD SCORE: BMI, AST/ALT > 0.8, DM

# CAN SERUM FERRITIN DIAGNOSE PRESENCE AND SEVERITY OF LIVER FIBROSIS?

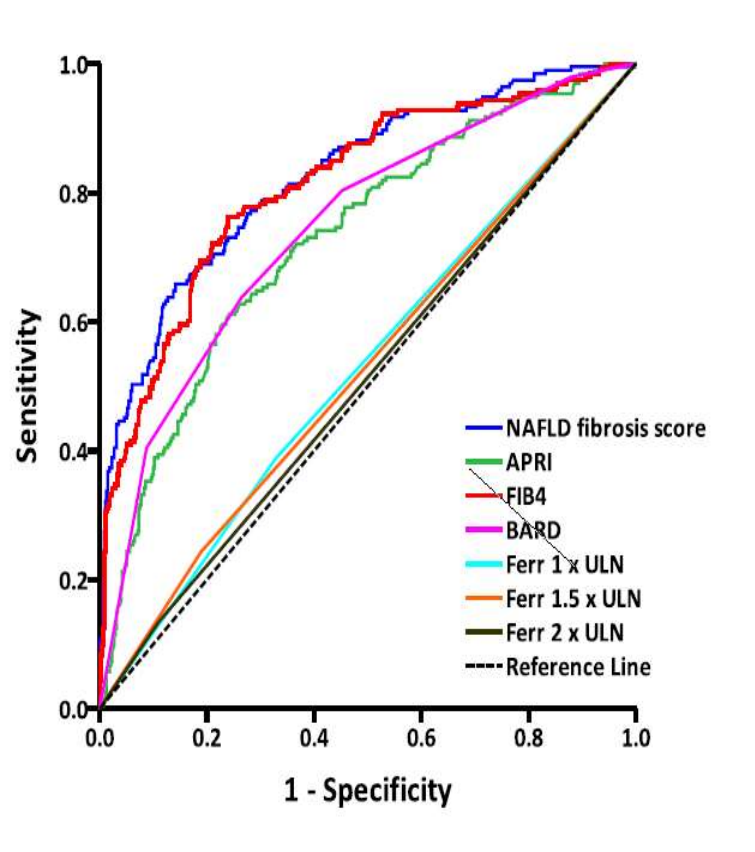
- INTERNATIONAL, RETROSPECTIVE COHORT STUDY OF 1014 PATIENTS WITH LIVER BIOPSY CONFIRMED NAFLD
- 55% OBESE, 31% OVERWEIGHT
- PREVALENCE OF SERUM FERRITIN >ULN WAS 33%
- FERRITIN >2 X ULN WAS 10%
- PATIENTS WITH ELEVATED SERUM FERRITIN WERE MORE LIKELY TO HAVE HISTOLOGICAL DIAGNOSIS OF DEFINITIVE NASH (45.9% S 34.8%, RESPECTIVELY, P<0.001), AND ADVANCED (STAGE 3-4) LIVER FIBROSIS (33.3% VS 23.5%, P = 0.001)

Angulo et al, CGH 2014

Diagnosis accuracy of simple scoring systems to differentiate between patients with and without advanced (stage 3–4) fibrosis

| | Area under the ROC curve (95% confidence intervals) [p value for the comparison with the score alone) | | | |
|---|---|---|---|---|
| **Score** | | ***Plus* Ferritin > ULN** | ***Plus* Ferritin > 1.5 x ULN** | ***Plus* Ferritin > 2 x ULN** |
| **NAFLD-FS** | 0.83 (0.79, 0.86) | 0.84 (0.80, 0.88) [p = 0.64] | 0.84 (0.80, 0.87) [p = 0.63] | 0.84 (0.80, 0.87) [p = 0.68] |
| **BARD** | 0.72 (0.69, 0.76) | 0.75 (0.72, 0.79) [p = 0.27] | 0.76 (0.72, 0.79) [p = 0.23] | 0.74 (0.70, 0.77) [p = 0.24] |
| **APRI** | 0.74 (0.70, 0.78) | 0.74 (0.70, 0.76) [p = 0.99] | 0.74 (0.70, 0.78) [p = 0.99] | 0.73 (0.69, 0.77) [p = 0.92] |
| **FIB-4** | 0.81 (0.78, 0.85) | 0.82 (0.78, 0.85) [p = 0.92] | 0.82 (0.78, 0.85) [p = 0.92] | 0.82 (0.78, 0.85) [p = 0.92] |

# DIAGNOSTIC ACCURACY TO DISTINGUISH BETWEEN PATIENTS WITH AND WITHOUT ADVANCED (STAGE 3-4) FIBROSIS



# IMPACT OF IRON REDUCTION ON INSULIN RESISTANCE, OXIDATIVE STRESS IN NAFLD

| Characteristics | Control Group (n= 41) | Venesection Group (n=31) | P value |
|---|---|---|---|
| Age (years) | 50.4 (11.8) | 53.1 (9.7) | 0.3 |
| Male sex | 24 (58%) | 20 (61%) | 0.9 |
| BMI (kg/m2) | 31.1 (5.3) | 31.8 (4.6) | 0.6 |
| Diabetes | 7 (17%) | 6 (18%) | 0.9 |
| Hypertension | 10 (24%) | 10 (30%) | 0.6 |
| Dyslipidemia | 6 (15%) | 5 (15%) | 0.6 |
| ALT (IU/L) | 48 )38-85) | 48 (37-86) | 0.9 |
| Triglyceride (mg/dL) | 133 (102-221) | 133 (106-173) | 0.4 |
| Ferritin (ng/mL) | 208 (229) | 255 (148) | 0.3 |
| Hepatic IC (mmol/kg) | 19.8 (9.5) | 23.0 (17.4) | 0.4 |
| Hepatic steatosis (%) | 17.4 (11.8) | 19.1 (9.7) | 0.5 |

Prospective Randomized Controlled Trial
N= 74, 6 month phlebotomy vs control

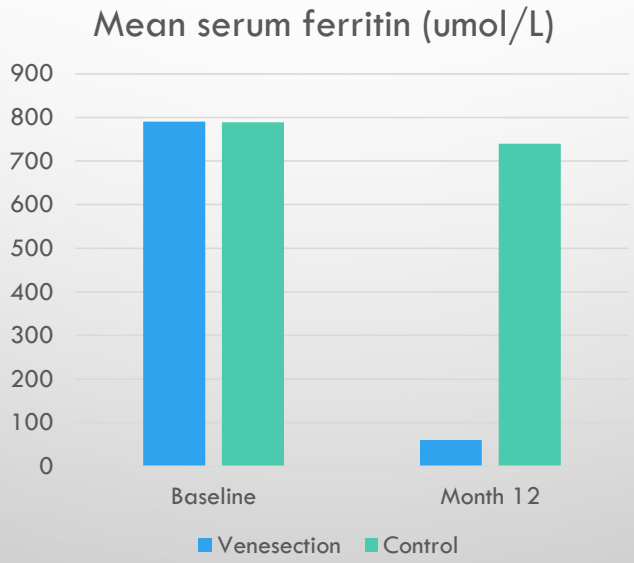
Adams et al. Hepatology 2015

- PHLEBOTOMY GROUP: MEDIAN 7 (1-19 SESSIONS)
- AT 6 MONTHS, THERE WAS NO DIFFERENCE IN HEPATIC STEATOSIS, ALT OR INSULIN RESISTANCE IN BOTH GROUPS

# VENESECTION IN DYSMETABOLIC IRON OVERLOAD SYNDROME

Randomized controlled trial
N=274, 12 month phlebotomy + lifestyle vs lifestyle alone

- **INCLUSION CRITERIA**
- LIVER IRON >50 UMOL/G
- 1 METABOLIC RISK FACTOR
- ABSENCE OF T2D, EXCESS ALCOHOL, CHRONIC INFLAMMATION, CHRONIC HEPATITIS, C282Y/C282Y
- **BASELINE CHARACTERISTICS**
- 85% MALE
- BMI 28
- MEAN FERRITIN 790
- MEAN HIC 88 UMOL/G




**No change** in
Insulin sensitivity
Glucose
Lipids
Liver enzymes

# HYPERFERRITINEMIA: CONCLUSIONS

- PRESENT IN 3-16% OF THE POPULATION
- VARIES SIGNIFICANTLY WITH ETHNICITY
- ASSOCIATED WITH GENETIC VARIANTS, OBESITY, FATTY LIVER, ALCOHOL AND INSULIN RESISTANCE
- RESPONDS TO LIFESTYLE INTERVENTION NOT VENESECTION

# CASE 1: CONTINUED

- A) HEMOCHROMATOSIS GENE TESTING
- B) CHECK HBA1C- 58 → STARTED ON JARDIANCE
- C) CHECK ETOH INTAKE
- D) LIVER BIOPSY → NO, LAST FIBROSCAN NOV 2020 6.6KPA
- E) VENESECTION

# CASE 1: PROGRESS

Previous results

| | 24/03/21 16:36 | 07/09/22 11:57 | 21/09/22 12:05 | 05/04/23 12:12 | 07/06/23 14:23 |
|---|---|---|---|---|---|
| Serum Iron | 25 | 31 | 31 | 36 | 24 |
| Transferrin | 2.8 | 2.9 | 3.0 | 2.6 | 2.9 |
| Transferrin Saturation | 0.36 | 0.43 | 0.41 | 0.55 | 0.33 |
| Ferritin | 2326 | 2738 | 3819 | 3000 | 1673 |
| Comment | | | | | |

| | 07/09/22 11:57 | 21/09/22 12:05 | 05/04/23 12:12 | 05/04/23 12:12 | 07/06/23 14:23 |
|---|---|---|---|---|---|
| Total Bilirubin | 10 | 13 | 10 | | 8 |
| Alk. Phosphatase | 165 | 128 | 151 | | 168 |
| GGT | 256 | 263 | 377 | | 284 |
| ALT | 107 | 133 | 102 | | 77 |
| Total Protein | 74 | 80 | 73 | | 77 |
| Albumin | 41 | 44 | 39 | 39 | 40 |
| Globulin | 33 | 36 | 34 | | 37 |
| Comment | | | | | |

# HEPATITIS B: SHOULD WE EXPAND CURRENT TREATMENT GUIDELINES?

# HEPATITIS B: CASE 1

- 40 YEAR OLD MALE
- HBEAG NEGATIVE
- HBV DNA 20,000 IU/ML
- LFTS: ALT 50

- DOES HE NEED ANTIVIRAL TREATMENT?

# HEPATITIS B: CASE 2

- 40 YEAR OLD CHINESE MALE
- HBEAG POSITIVE
- NORMAL ALT OF 25
- HBV DNA >8 LOGS

- SHOULD HE BE STARTED ON ANTIVIRAL TREATMENT?

# HEPATITIS B: CASE 3

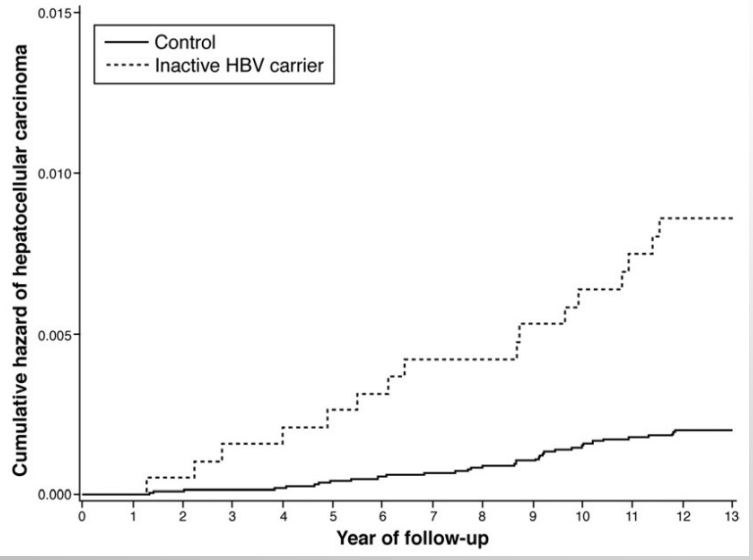
- 60 YEAR OLD FEMALE
- HBEAG NEGATIVE
- ALT NORMAL 25
- HBV DNA 10,000IU/ML

- DOES SHE NEED ANTIVIRAL TREATMENT?

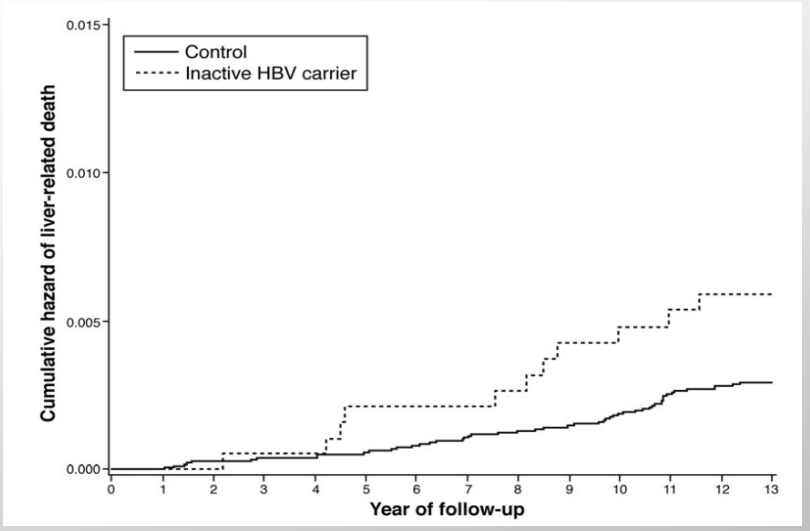
# LOWER HBV DNA LEVELS ALSO ASSOCIATED WITH LIVER COMPLICATIONS

**Progression to HCC**
**(p<0.001 by log-rank test)**

Inactive carriers defined as HBV DNA <2000IU/mL and ALT <45 U/L



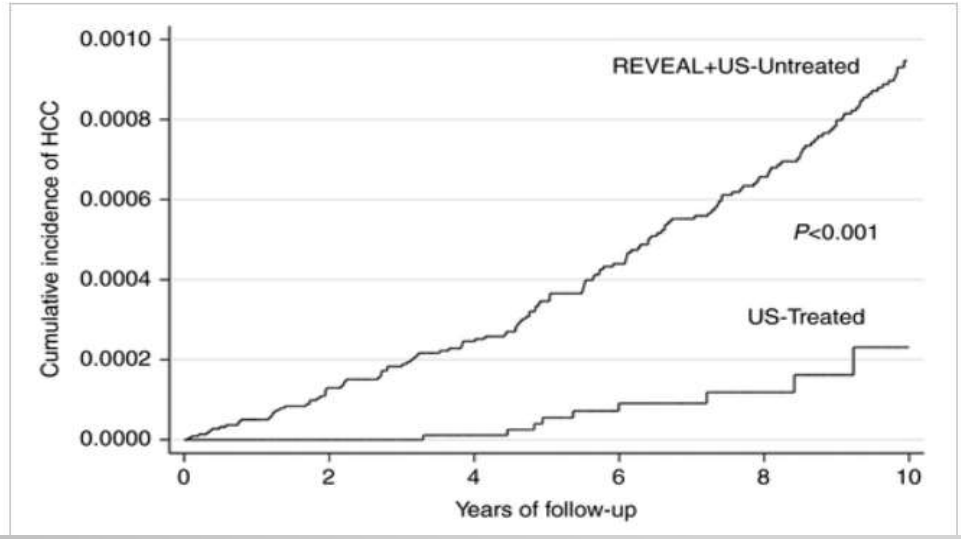

**Progression to liver-related complications**
**(p=0.029 by log rank test)**




Chen JD. Gastroenterology 2010; 138: 1747-54.

# HBV DNA SUPPRESSION ASSOCIATED WITH DECREASED HCC RISK

**973 CHB pts on long term NUCs matched with 4936 untreated controls**
**- Hazard of developing HCC adjusted by REACH-B score**




**261 HCC (5.5%)**

**12 HCC (1.2%)**

**Adjusted HR 0.3 (0.13-0.43) P<0.0001**

Lin D et al. AP&T; 44(8): 846-55.

# HBV SUPPRESSION WILL REDUCE HBV INTEGRATION

- BIOPSIES AT BASELINE, AFTER 1 YEAR, 10 YEARS NUC
- → HEPATOCYTE DNA EXTRACTED, DIGESTED AND CLONED
- 100% HAD INTEGRATION AT ESP CHR 16,17,21,22
- 1/3 HAD CHROMOSOMAL TRANSLOCATION
- → AFTER 10 YEARS, INTEGRATION CLONES REDUCED BY >99.9% AND 60% BIOPSIES HAD NO DETECTABLE INTEGRATION

Chow N et al. Hepatology 2020; 72: 19A.

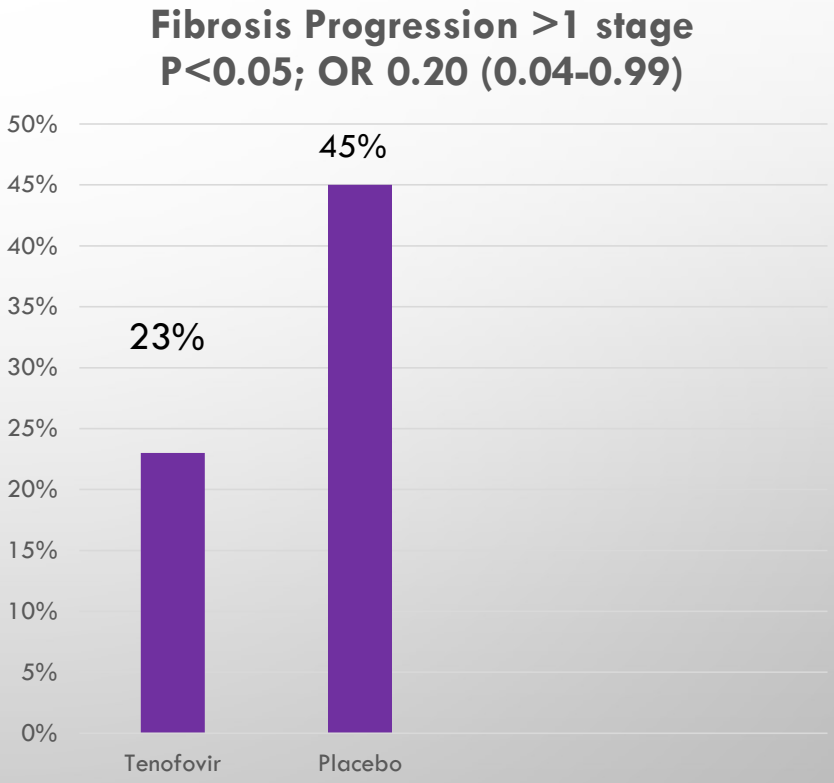
# HBV SUPPRESSION PREVENTS CIRRHOSIS IN PATIENTS WITH LOW ALT

160 Taiwanese patients with ALT <2xULN + HBV DNA >2000 IU/mL

- Randomised 1:1 to tenofovir or placebo
- Biopsied at baseline and after 3 years

- Progression to cirrhosis : 3% vs 14% (P<0.01; OR 0.37 (0.17-0.78)

ALT is a poor predictor of fibrosis progression in CHB → Treat all eAg neg CHB pts with DNA >2000?




Hsu YC et al. Lancet Inf Dis 2021;21(6):823-33.

# UNTREATED "IMMUNE TOLERANT" PATIENTS DEVELOP COMPLICATIONS

- 413 UNTREATED "IMMUNOTOLERANT PATIENTS WITH HBV DNA >5 LOG, ALT <ULN (19 IN FEMALES, 30 IN MALES), NO FIBROSIS
- MATCHED WITH 1497 TREATED PATIENTS WITH IMMUNE ACTIVE CHB (BASELINE ALT >2X ULN)
- IT GROUP SIGNIFICANTLY YOUNGER THAN IA GROUP (MEAN AGE 38 VS 40 YRS AT BASELINE, P = 0.04)
- 10 YEAR CUMULATIVE INCIDENCE OF HCC (12.7% VS 6.1%, P =0.001)
- DEATH/TRANSPLANTATION (9.7% VS 3.4%, P<0.001)

Kim GA et al. Gut 2018; 67(5):945-52.

# NEW EASL TREATMENT CRITERIA

- PATIENTS WITH HBV DNA >20,000IU/ML AND ALT >2X ULN SHOULD START TREATMENT REGARDLESS OF THE DEGREE OF FIBROSIS
- PATIENTS WITH HBEAG POSITIVE CHRONIC HBV DEFINED BY PERSISTENTLY NORMAL ALT AND HIGH HBV DNA LEVELS, MAY BE TREATED IF THEY ARE OLDER THAN 30 YEARS OF AGE, REGARDLESS OF THE SEVERITY OF FIBROSIS

# REACH B HCC SCORE

- GENDER: FEMALE (0), MALE (+2)
- AGE, YEARS: 30-34 (0), 35-39 (+1), 40-44 (+2), 45-49 (+3), 50-54 (+4), 55-59 (+5), 60-65 (+6)
- ALT (U/L) : <15 (0), 15-44 (+1), >45 (+2)
- HBEAG : NEGATIVE (0), POSITIVE (+2)
- HBV DNA (COPIES/ML): <300 (0), 300-999 (0), 10,000-99,999 (+3),
  - 100,000-999,999 (+5), >10X6 (+4)

# CASE 1

- 40 YEAR OLD MALE
- HBEAG NEGATIVE
- HBV DNA 20,000 IU/ML
- LFTS: ALT 50

- REACH B HCC SCORE = 11 POINTS (8.4% 10 YEAR RISK OF HCC)

# CASE 2

- 40 YEAR OLD CHINESE MALE
- HBEAG POSITIVE
- NORMAL ALT OF 30
- HBV DNA >8 LOGS

- REACH B HCC SCORE = 11 POINTS (8.4% 10 YEAR RISK OF HCC)

# CASE 3

- 60 YEAR OLD FEMALE
- HBEAG NEGATIVE
- ALT NORMAL 25
- HBV DNA 100,000IU/ML

- REACH B HCC SCORE = 12 POINTS (13.4% 10 YEAR RISK OF HCC)

# IMPACT ON DISEASE BURDEN IF WE TREAT EVERYONE WHO IS HBSAG+ AND HBV DNA >2000

- COMPARED TO THE BASE SCENARIO, THIS STRATEGY WOULD:
- 1) PREVENT 2000 CASES OF DECOMPENSATED CIRRHOSIS
- 2) PREVENT 8500 CASES OF HCC
- 3) AND SAVE ALMOST 10,000 LIVES THROUGH TO 2050
- TREATING ALL HBV DNA >2000 COST-EFFECTIVE IF PER PATIENT COST IS USD 2000/YR AND WOULD BE COST SAVING AT <USD750/YR
- IN 2023 IN NZ, GENERIC NUCS COST USD 300-400/YR

# CHANGING LANDSCAPE IN HBV MANAGEMENT IN 2023

- HBV DNA >2000IU/ML LEADS TO WORSE OUTCOMES
- ALT IS NOT A SIGNIFICANT INDICATOR OF RISK
- THE LONGER YOU LET THE VIRUS REPLICATE THE MORE INTEGRATION EVENTS TAKE PLACE, THE HIGHER THE RISK OF HCC
- HBV SUPPRESSION IS THE BEST FORM OF CANCER PREVENTION
- TREAT THE VIRUS JUST LIKE WE DO FOR HIV
- GENERICS HAVE MADE THIS HIGHLY COST SAVING
- MANY MORE PATIENTS SHOULD BE STARTED ON ANTIVIRAL TREATMENT

# SAFETY AND EFFICACY OF VIR-2218 WITH OR WITHOUT PEGYLATED INTERFERON ALFA IN VIRALLY-SUPPRESSED PARTICIPANTS WITH CHRONIC HEPATITIS B VIRUS INFECTION: POST-TREATMENT FOLLOW UP

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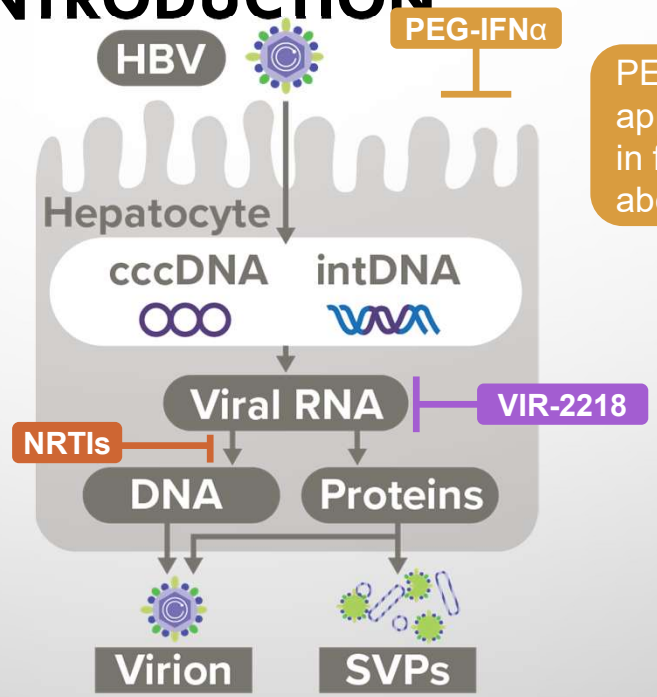
# INTRODUCTION

- Preliminary data have shown that combining VIR-2218 and PEG-IFNα results in deeper HBsAg declines compared with VIR-2218 alone[1]

- We hypothesize that lowering HBsAg with VIR-2218 in the context of immune stimulation by PEG-IFNα may lead to HBsAg seroclearance in a greater proportion of patients



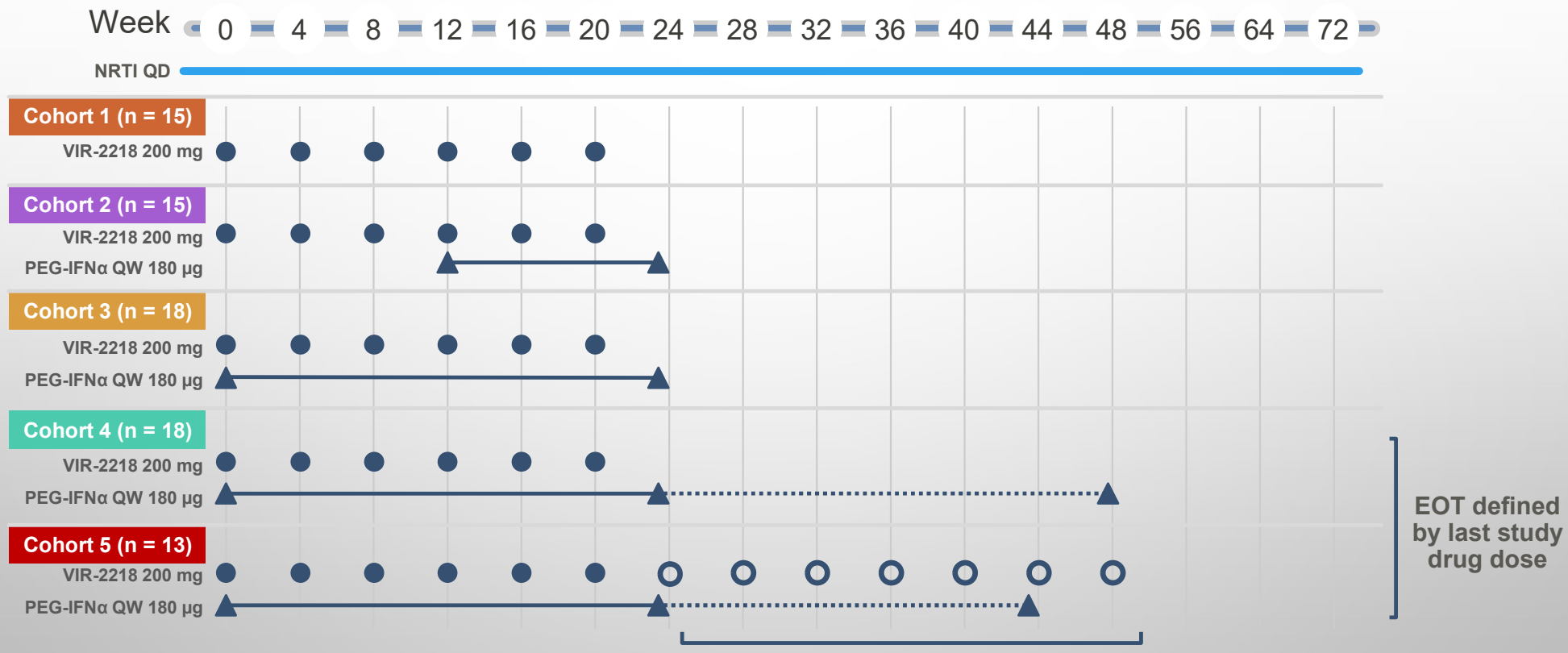

PEG-IFNα is the only approved therapy to result in functional cure, but in only about 3% to 7% of patients[2]

VIR-2218 is a GalNAc-conjugated ESC+ siRNA targeting the HBx region of HBV genome that reduces HBsAg in patients with chronic HBV infection[3]

**1.** Yuen MF, et al. Hepatology 2022; 76:S18; **2.** EASL. *J Hepatol.* 2017;67:370-398; **3.** Gane E, et al. J Hepatol 2021;75(2): S287.

**Abbreviations:** EASL, European Association for the Study of the Liver; cccDNA, covalently closed circular DNA; intDNA, integrated DNA; ESC+, enhanced stabilization chemistry plus; GalNAc, trivalent N-acetylgalactosamine; HBsAg, hepatitis B surface antigen; HBV, hepatitis B virus; HBx, hepatitis B virus X protein; PEG-IFNα, pegylated interferon alfa-2a; RNA, ribonucleic acid; siRNA, small interfering RNA; SVPs, subviral particles.

# A PHASE 2 TRIAL EVALUATING VIR-2218 WITH AND WITHOUT PEG-IFNA




[a]HBsAg assay LLOQ and LLOD are 0.05 IU/mL.

**Abbreviations:** EOT, end of treatment; HBsAg, hepatitis B surface antigen; LLOD, lower limit of detection; LLOQ, lower limit of quantitation; NRTI, nucleos(t)ide reverse transcriptase inhibitor; PEG-IFNα, pegylated interferon alfa-2a; QD, daily; QW, every week; anti-HBs, hepatitis B surface antibody.

# KEY INCLUSION/EXCLUSION CRITERIA

## Inclusion

- Age 18 to 65 years (inclusive)
- Chronic HBV infection defined as positive serum HBsAg for ≥6 months
- On NRTI therapy for ≥2 months
- HBsAg >50 IU/mL
- HBV DNA <90 IU/mL

## Exclusion

- Significant fibrosis or cirrhosis (FibroScan >8.5 kPa at screening or Metavir F3/F4 liver biopsy within 1 year)
- Bilirubin, INR, or prothrombin time >ULN
- ALT or AST >2 x ULN
- Active HIV, HCV, or HDV infection

**Abbreviations:** ALT, alanine aminotransferase; AST, aspartate aminotransferase; DNA, deoxyribonucleic acid; HBsAg, hepatitis B surface antigen;
HBV, hepatitis B virus; HCV, hepatitis C virus; HDV, hepatitis D Virus; HIV, human immunodeficiency virus; INR, international normalised ratio;
NRTI, nucleos(t)ide reverse transcriptase inhibitor; ULN, upper limit of normal.

# DEMOGRAPHICS AND BASELINE CHARACTERISTICS

| Participants | Cohort 1 (n = 15) | Cohort 2 (n = 15) | Cohort 3 (n = 18) | Cohort 4 (n = 18) | Cohort 5 (n = 13) |
|---|---|---|---|---|---|
| | VIR-2218 x 6 | VIR-2218 x 6 lead-in + PEG-IFNα x 12 | VIR-2218 x 6 + PEG-IFNα x 24 | VIR-2218 x 6 + PEG-IFNα x ≤48 | VIR-2218 x 13 + PEG-IFNα x ≤44 |
| **HBeAg-positive, n (%)** | 4 (26.7) | 6 (40.0) | 7 (38.9) | 6 (33.3) | 3 (23.1) |
| **Age (years), mean (SD)** | 50.3 (8.6) | 46.6 (7.8) | 48.7 (5.8) | 45.2 (9.4) | 48.5 (7.6) |
| **Male (sex), n (%)** | 13 (86.7) | 13 (86.7) | 14 (77.8) | 15 (83.3) | 7 (53.8) |
| **Race, n (%)** | | | | | |
| Asian | 12 (80.0) | 13 (86.7) | 16 (88.9) | 18 (100.0) | 13 (100.0) |
| White | 0 | 0 | 1 (5.6) | 0 | 0 |
| Other | 3 (20.0) | 2 (13.3) | 1 (5.6) | 0 | 0 |
| **HBsAg ($log_{10}$ IU/mL), median (range)** | 3.4 (2.6, 4.1) | 3.2 (2.2, 4.0) | 3.4 (2.2, 4.2) | 2.9 (1.9, 4.3) | 3.7 (2.1, 4.4) |
| **ALT (U/L), mean (SD)** | 21.5 (10.1) | 25.0 (12.4) | 21.7 (12.0) | 19.7 (7.1) | 22.6 (10.1) |
| **ALT >ULN, n (%)** | 1 (6.7) | 1 (6.7) | 1 (5.6) | 0 | 1 (7.7) |

**Abbreviations:** ALT, alanine aminotransferase; HBeAg, hepatitis B e antigen; HBsAg, hepatitis B surface antigen; PEG-IFNα, pegylated interferon alfa-2a; SD, standard deviation; ULN, upper limit of normal.

# VIR-2218 WITH OR WITHOUT PEG-IFNα

| Participants, n (%) | Cohort 1 (n = 15) | Cohort 2 (n = 15) | Cohort 3 (n = 18) | Cohort 4 (n = 18) | Cohort 5 (n = 13) |
|---|---|---|---|---|---|
| | VIR-2218 x 6 | VIR-2218 x 6 lead-in + PEG-IFNα x 12 | VIR-2218 x 6 + PEG-IFNα x 24 | VIR-2218 x 6 + PEG-IFNα x ≤48 | VIR-2218 x 13 + PEG-IFNα x ≤44 |
| **Any TEAEs[a]** | 9 (60.0) | 13 (86.7) | 16 (88.9) | 17 (94.4) | 13 (100.0) |
| Grade 1 | 7 (46.7) | 9 (60.0) | 7 (38.9) | 10 (55.6) | 4 (30.8) |
| Grade 2 | 2 (13.3) | 4 (26.7) | 7 (38.9) | 4 (22.2) | 6 (46.2) |
| Grade 3 | 0 | 0 | 2 (11.1) | 2 (11.1) | 3 (23.1) |
| Grade 4 | 0 | 0 | 0 | 1 (5.6) | 0 |
| **Treatment-related TEAEs[a]** | 3 (20.0) | 12 (80.0) | 13 (72.2) | 15 (83.3) | 13 (100.0) |
| Related to VIR-2218 | 3 (20.0) | 4 (26.7) | 8 (44.4) | 7 (38.9) | 6 (46.2) |
| Related to PEG-IFNα | N/A | 12 (80.0) | 12 (66.7) | 14 (77.8) | 13 (100.0) |
| Related to VIR-2218 and PEG-IFNα | N/A | 1 (6.7) | 5 (27.8) | 3 (16.7) | 4 (30.8) |
| **SAE[b]** | 0 | 0 | 1 (5.6) | 1 (5.6) | 1 (7.7) |
| **Study discontinuation due to TEAE** | 0 | 0 | 0 | 0 | 0 |

- Most TEAEs were consistent with the known effects of PEG-IFNα
- No SAEs were related to VIR-2218

[a]TEAE is defined as any AE with onset after study drug start and within 30 days of the last dose of study drug.
[b]3 SAEs have been reported: ankle fracture (Cohort 3, n = 1), gall bladder pain (Cohort 5, n = 1) unrelated to study treatments, and mania (Cohort 4, n = 1) related to PEG-IFNα.
**Abbreviations:** AE, adverse event; N/A, not applicable; PEG-IFNα, pegylated interferon alfa-2a; SAE, serious adverse event; TEAE, treatment-emergent adverse event.

# EFFECTS ON ALT, NEUTROPHIL, AND

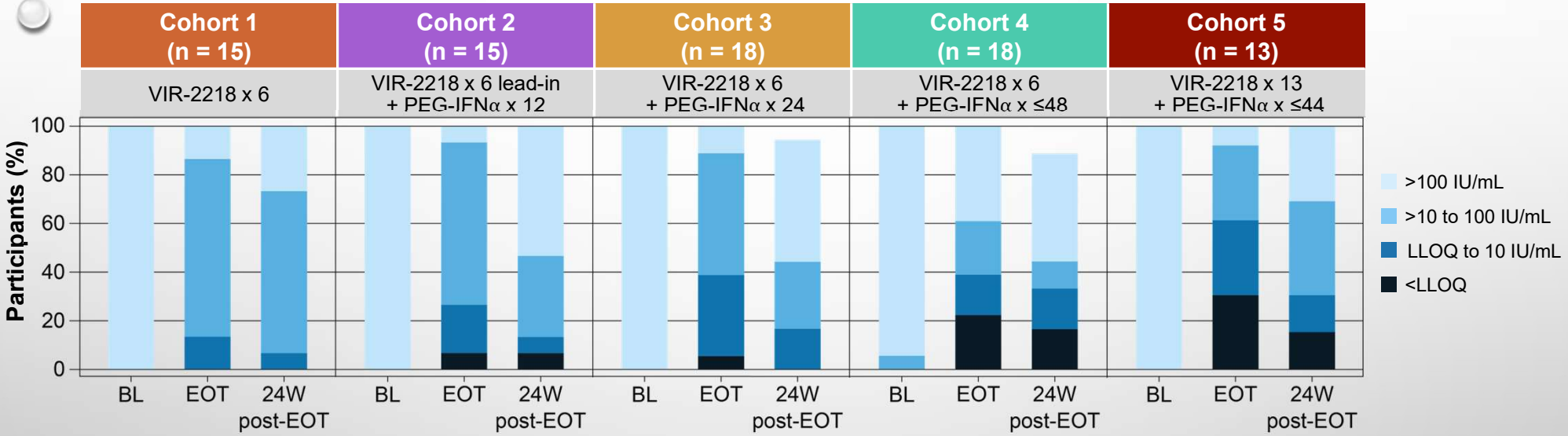
| Participants, n (%) | Cohort 1 (n = 15) | Cohort 2 (n = 15) | Cohort 3 (n = 18) | Cohort 4 (n = 18) | Cohort 5 (n = 13) |
|---|---|---|---|---|---|
| | VIR-2218 x 6 | VIR-2218 x 6 lead-in + PEG-IFNα x 12 | VIR-2218 x 6 + PEG-IFNα x 24 | VIR-2218 x 6 + PEG-IFNα x ≤48 | VIR-2218 x 13 + PEG-IFNα x ≤44 |
| **ALT level increase** | | | | | |
| Grade 1 | 2 (13.3) | 12 (80.0) | 12 (66.7) | 14 (77.8) | 9 (69.2) |
| Grade 2 | 0 | 1 (6.7) | 2 (11.1) | 2 (11.1) | 2 (15.4) |
| Grade 3 | 0 | 0 | 1 (5.6) | 1 (5.6) | 0 |
| **Neutrophil level decrease** | | | | | |
| Grade 1 | 3 (20.0) | 4 (26.7) | 4 (22.2) | 1 (5.6) | 1 (7.7) |
| Grade 2 | 1 (6.7) | 8 (53.3) | 10 (55.6) | 6 (33.3) | 6 (46.2) |
| Grade 3 | 0 | 2 (13.3) | 3 (16.7) | 10 (55.6) | 6 (46.2) |
| Grade 4 | 0 | 0 | 0 | 1 (5.6) | 0 |
| **Platelet level decrease** | | | | | |
| Grade 1 | 1 (6.7) | 10 (66.7) | 10 (55.6) | 14 (77.8) | 9 (69.2) |

- Majority of ALT elevations resolved within 24 weeks post-EOT
- Majority of neutrophil and platelet abnormalities resolved within 4 weeks post-EOT[a]

Grading defined by CTCAE

**Abbreviations:** ALT, alanine transaminase; CTCAE, Common Terminology Criteria for Adverse Events; PEG-IFNα, pegylated interferon alfa-2a.

# LONGER DURATION OF COMBINATION TREATMENT RESULTED IN GREATER HBSAG DECLINE AND RATES OF SEROCLEARANCE




- HBsAg seroclearance[a] was observed only in participants receiving the combination of VIR-2218 and PEG-IFNα
- Compared with other cohorts, more participants (62% [8/13]) in Cohort 5 achieved HBsAg levels <10 IU/mL at EOT
  - 69% (9/13) of participants sustained HBsAg levels <100 IU/mL at 24-weeks post-EOT

[a]Seroclearance defined as HBsAg <0.05 IU/mL (LLOQ).
**Abbreviations:** BL, baseline; EOT, end of treatment; HBsAg, hepatitis B surface antigen; LLOQ, lower limit of quantitation; PEG-IFNα, pegylated interferon alfa-2a; W, weeks.

# LONGER TREATMENT DURATIONS WERE ASSOCIATED WITH HIGHER RATES OF HBSAG SEROCLEARANCE

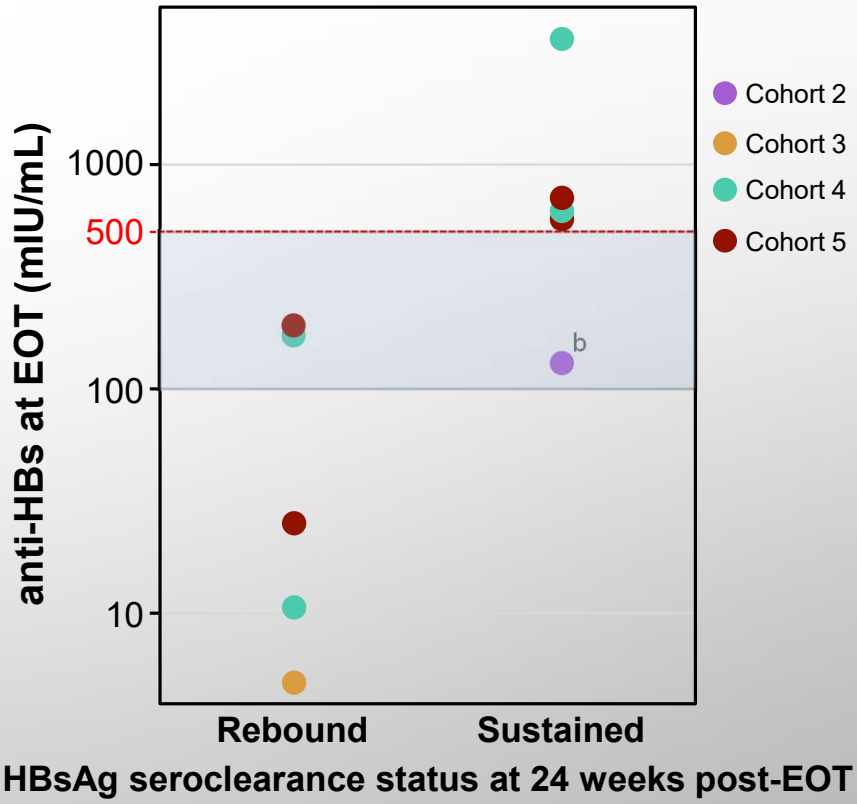
| Participants with HBsAg seroclearance, n (%) | Cohort 1 (n = 15) | Cohort 2 (n = 15) | Cohort 3 (n = 18) | Cohort 4 (n = 18) | Cohort 5 (n = 13) |
|---|---|---|---|---|---|
| | VIR-2218 x 6 | VIR-2218 x 6 lead-in + PEG-IFNα x 12 | VIR-2218 x 6 + PEG-IFNα x 24 | VIR-2218 x 6 + PEG-IFNα x ≤48 | VIR-2218 x 13 + PEG-IFNα x ≤44 |
| At EOT[a] | 0 (0) | 1 (6.7) | 1 (5.6) | 4 (22.2) | 4 (30.8) |
| **At 24 weeks post-EOT** | 0 (0) | 1 (6.7) | 0 (0) | 3[b] (16.7) | 2 (15.4) |
| HBsAg at baseline | | | | | |
| <1,000 IU/mL | 0/3 (0) | 1/5 (20) | 0/6 (0) | 2/10 (20) | 1/5 (20) |
| >1000 IU/mL | 0/12 (0) | 0/10 (0) | 0/12 (0) | 1/8 (12.5) | 1/8 (12.5) |
| HBeAg at baseline | | | | | |
| HBeAg-positive | 0/4 (0) | 0/6 (0) | 0/7 (0) | 1/6 (16.7) | 1/3 (33.3) |
| HBeAg-negative | 0/11 (0) | 1/9 (11.1) | 0/11 (0) | 2/12 (16.7) | 1/10 (10) |

- AMONG 31 PARTICIPANTS RECEIVING 48-WEEK REGIMENS OF VIR-2218 AND PEG-IFNα
  - 8 (25.8%) HAD HBSAG SEROCLEARANCE AT EOT
  - 5 (16.1%) SUSTAINED HBSAG SEROCLEARANCE AT 24 WEEKS POST-EOT
- ALL PARTICIPANTS WHO HAD HBSAG REBOUNDS AFTER SEROCLEARANCE MAINTAINED LEVELS <100 IU/ML AT 24 WEEKS POST-EOT

[a]End of treatment refers to the last day of study drug administration; in cohorts 4 and 5 some participants met efficacy criteria to stop treatment early.
[b]Two participants with HBsAg seroclearance at EOT had rebounds; an additional participant had HBsAg seroclearance after EOT but maintained it through 24 weeks post-EOT. Seroclearance defined as HBsAg <0.05 IU/mL (LLOQ).
**Abbreviations:** EOT, end of treatment; HBeAg, hepatitis B e antigen; HBsAg, hepatitis B surface antigen; LLOQ, lower limit of quantitation; PEG-IFNα, pegylated interferon alfa-2a.

# HIGHER ANTI-HBS TITER AT EOT PREDICTS HBSAG SEROCLEARANCE DURABILITY

- AMONG PARTICIPANTS WHO HAD HBSAG SEROCLEARANCE BY EOT[A]:
  - ALL PARTICIPANTS (4/4) WITH ANTI-HBS LEVELS >500 MIU/ML AT EOT HAD SUSTAINED HBSAG SEROCLEARANCE AT 24 WEEKS POST-EOT
  - ALL PARTICIPANTS (3/3) WITH ANTI-HBS <100 MIU/ML AT EOT EXPERIENCED A REBOUND IN HBSAG
  - THREE PARTICIPANTS HAD ANTI-HBS LEVELS BETWEEN 100–500 MIU/ML; 2 EXPERIENCED A REBOUND, AND 1 SUSTAINED HBSAG SEROCLEARANCE THROUGH 24 WEEKS POST-EOT




[a] One participant had HBsAg seroclearance after EOT and is not included in the analyses.
[b] Participant had anti-HBs >1,500 mIU/mL 4 weeks after EOT.
Seroclearance defined as HBsAg <0.05 IU/mL (LLOQ).
**Abbreviations:** EOT, end of treatment; anti-HBs, hepatitis B surface antibody; HBsAg, hepatitis B surface antigen; LLOQ, lower limit of quantitation.

# SUMMARY OF RESULTS

- VIR-2218 ALONE AND IN COMBINATION WITH PEG-IFNα WAS GENERALLY WELL TOLERATED
  - MOST ADVERSE EVENTS WERE AS EXPECTED WITH PEG-IFNα AND RESOLVED AFTER EOT
  - NO PARTICIPANTS DISCONTINUED THE STUDY DUE TO TEAES
- AMONG PARTICIPANTS RECEIVING 48 WEEKS OF CONCURRENT VIR-2218 PLUS PEG-IFNα:
  - 31% (4/13) HAD HBSAG SEROCLEARANCE AT EOT
  - 15% (2/13) HAD SUSTAINED HBSAG SEROCLEARANCE FOR 24 WEEKS POST-EOT
- LONGER DURATIONS OF TREATMENT WITH BOTH VIR-2218 AND PEG-IFNα WERE ASSOCIATED WITH GREATER HBSAG DECLINE AND A HIGHER INCIDENCE OF HBSAG SEROCLEARANCE
- ANTI-HBS TITERS >500 MIU/ML AT EOT WERE ASSOCIATED WITH SUSTAINED HBSAG SEROCLEARANCE AT 24 WEEKS POST-EOT
- ADDITIONAL FOLLOW UP IS ONGOING

Seroclearance defined as HBsAg <0.05 IU/mL (LLOQ).

**Abbreviations:** EOT, end of treatment; anti-HBs, hepatitis B surface antibody; HBsAg, hepatitis B surface antigen; LLOQ, lower limit of quantitation; PEG-IFNα, pegylated interferon alfa-2a; TEAE, treatment-emergent adverse event.

# BEPIROVIRSEN (GSK)

- PHASE 2B TRIAL (B-CLEAR)
- 12 OR 24 WEEKS TREATMENT WITH BEPI IN CHB PATIENTS ON STABLE NA TREATMENT OR NOT ON NA
- PRIMARY ENDPOINTS = PROPORTION OF PATIENTS ACHIEVING HBSAG LEVELS <LLOQ AND HBV DNA LEVELS <LLOQ SUSTAINED FOR 24 WEEKS AFTER END OF TREATMENT

# BEPIROVIRSEN

- 2 PARALLEL COHORTS, ONE FOR PATIENTS ON NA AND ONE FOR THOSE NOT ON NA
- PATIENTS FROM EACH ARM RANDOMIZED TO ONE OF 4 TREATMENT ARMS
- TREATMENT ADMINISTERED WITH OR WITHOUT LOADING DOSES (LD) ON DAYS 4 AND 11
- - BEPIROVIRSEN 300MG WITH LD FOR 24 WEEKS
- - BEPIROVIRSEN 300MG WITH LD FOR 12 WEEKS THEN 150MG FOR 12 WEEKS
- - BEPIROVIRSEN 300MG WITH LD FOR 12 WEEKS THEN PLACEBO FOR 12 WEEKS
- - PLACEBO WITH LD FOR 12 WEEKS THEN BEPIROVIRSEN 300MG WITHOUT LD FOR 12 WKS

# BEPIROVIRSEN (RESULTS)

- PATIENTS ON NA TREATMENT (N=227)
- 24 WKS TREATMENT OF 300MG BEPIROVIRSEN RESULTED IN HBSAG <LLOQ AND HBV DNA <LLOQ IN 28% PATIENTS AT END OF TREATMENT

- FOR PATIENTS NOT ON NA (N=230)
- 24 WKS TREATMENT OF 300MG BEPIROVIRSEN RESULTED IN HBSAG <LLOQ AND HBV DNA <LLOQ IN 29% PATIENTS AT END OF TREATMENT
- TREATMENT RELATED SAES 1% IN EACH GROUP
- BY 24 WKS AFTER TREATMENT, SUSTAINED HBSAG RESPONSE IN 25%
- PHASE III TRIAL RECRUITING NOW