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ABSTRACT BOOK

HBV ENDPOINTS

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ePOSTER ABSTRACT PRESENTATIONS

OP-01 A novel pyrazole HBV nucleocapsid formation inhibitor demonstrating high activity against HBV variants that are resistant to class I and class II core protein allosteric modulators

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Background and aims: Current drugs for chronic hepatitis B (NAs and IFNα) rarely provide a cure for patients. New classes of drugs that hit different virus targets are welcomed to bring a higher cure rate in finite treatment periods. We have discovered a novel series of pyrazole compounds as HBV nucleocapsid formation inhibitors (represented by CB-HBV-001). CB-HBV-001 is structurally distinctive from the current class I (heteroaryldihydropyrimidines, e.g., BAY-41-4109 and GLS4) and class II (phenylpropenamides and sulfamoylbenzamides, e.g., AT-130 and NVR3-778) HBV core protein allosteric modulators (CpAMs). We evaluated the antiviral activities of CB-HBV-001, class I, and class II compounds against a panel of viruses containing different mutations in HBV core protein including the core T109I mutant that has shown resistance to both NV-010-001 and BAY 41-4109 core protein modulators (Klumpp et al., PNAS 2015).
Method: Virus infection of PHH and Southern Blot analysis were used to quantify the HBV cccDNA. HepG2 cells were transfected with 1.3x HBV DNA carrying different core mutations. Newly synthesized virus DNA was quantified by PCR.
Results: CB-HBV-001 accelerated the formation of capsid devoid of HBV pgRNA and DNA in a dose-dependent manner. When added at the time of virus infection of PHH, CB-HBV-001 inhibited HBV cccDNA formation and HBsAg level. In addition, when used in combination with IFN-alpha in PHH assay, CB-HBV-001 demonstrated an additive effect in inhibiting both HBsAg and DNA, while it only showed an additive inhibitory effect with Tenofovir on HBV DNA. Most importantly, CB-HBV-001 was highly active against a panel of viruses that contain mutations in core protein and that are resistant to classes I and II CpAMs.
Conclusion: The unique structure of CB-HBV-001 and its distinctive antiviral profile against core protein mutants distinguish itself from reported class I and class II HBV CpAMs. These characteristics of CB-HBV-001 warrant its clinic evaluation for treatment of chronic HBV infection.
Figure:

Mutation in HBV core protein	EC50(nM) (and Folds of Change in EC50 Value)				
		Class II		Class I	NA
	CB-HBV-001	Compound 2 (WO2017/181141)	AT-130	GLS4	ETV
F23Y	189 (6.1)	453 (12)	7573 (37)	38 (3.5)	2.7 (1.0)
L30F	59 (1.9)	228 (5.9)	1289 (6.3)	56 (5.1)	2.8 (1.0)
I105F	5.7 (0.2)	491 (13)	631 (3.1)	44 (4.0)	3.4 (1.2)
T33Q	159 (5.1)	>10000 (>257)	>10000 (>49)	428 (39)	2.8 (1.0)
T109I	23 (0.8)	10 (0.3)	81 (0.4)	134 (12.3)	1.4 (0.5)
U95551(wild type)	31 (1.0)	39 (1.0)	206 (1.0)	11 (1.0)	2.8 (1.0)

OP-02 Establishment of High Rates of Functional Cure of HBeAg Negative Chronic HBV Infection with REP 2139-Mg Based Combination Therapy: Ongoing Follow-up Results from the REP 401 Study

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Background and aims: The REP 401 study (NCT02565719) is assessing the safety and efficacy of REP 2139-Mg (clinical lead) or REP 2165-Mg combined with tenofovir disoproxil fumarate (TDF) and pegylated interferon α-2a (pegIFN) in Caucasian patients with HBeAg negative chronic HBV infection.
Method: Lead-in TDF therapy in 40 patients was followed by randomization into an experimental group (48 weeks of TDF, pegIFN and REP 2139-Mg or REP 2165-Mg) or an adaptive control group (24 weeks of TDF + pegIFN followed by cross over to 48 weeks of experimental therapy). All patients were subsequently entered into a treatment-free follow-up scheduled for 48 weeks. Viremia is monitored on the Abbott Architect and Realtime platforms.
Results: Baseline HBsAg was >1000 IU/ml in all patients and 14775.7 ± 9302 and 9018 ± 8743 IU/ml in adaptive control and experimental groups. Therapy was well tolerated except for one withdrawal due to pegIFN-related depression. Two additional participants withdrew where therapy was well tolerated. Antiviral responses between REP 2139-Mg and REP 2165-Mg were indistinguishable and not related to HBV genotype or HBsAg, ALT or fibrosis (Fibroscan) at baseline. Following the introduction of TDF, HBV DNA was well controlled in all patients during therapy. Following the introduction of NAPs, HBsAg reduction was >1 log in 36/40 patients and became <1 IU/ml in 28/40 and <0.05 IU/ml in 24/40 participants. Transaminase flares occurred in 38/40 patients and were correlated with reductions in HBsAg, were self-resolving during therapy and not accompanied by any evidence of liver dysfunction. Transaminase flares were especially pronounced (400-1748 U/L) in patients where HBsAg became <1 IU/ml and were accompanied by profound elevations in anti-HBs (up to 255, 055 mIU/ml).
As of the date of submission, treatment-free follow-up has been extended to 24 or 48 weeks in 34/40 patients completing treatment. Persistent and stable inactive chronic HBV (HBV DNA <2000 IU/ml with normal ALT) is present in 15/34 (44%) of participants. An additional 14/34 (41%) participants have functional cure (HBsAg and HBV DNA target not detected). Liver function has normalized in 94% of patients (versus 47% at baseline) and median hepatic stiffness consistent with F0 (≤7kPa) is present in 81% of patients (versus 52% at baseline).
Conclusion: A finite REP 2139-Mg based combination therapy with TDF and pegIFN is well tolerated and results in a high proportion of patients achieving control of infection not requiring further therapy under current guidelines. Transaminase flares appear therapeutic in nature and may reflect an immune mediated clearance of infected hepatocytes essential in establishing persistent control of chronic HBV infection.

OP-03 Pre-genomic HBV RNA and HBcrAg play important role in the predicting clinical outcomes in chronic hepatitis B patients suppressed on antiviral therapy with nucleos (t)ide analogues

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Background and aims: A dichotomous separation of HBV DNA and HBsAg concentrations occurs during natural history and treatment of chronic hepatitis B (CHB). HBsAg production originates from dual sources: cccDNA and integrated DNA. Serum pre-genomic (pg) HBV RNA and HBcrAg reflect cccDNA transcription during HBV replication and thus could prove useful clinical surrogates of silencing of cccDNA. We have evaluated the role of pgHBV RNA and HBcrAg in 3 independent cohorts of CHB patients and ability of these markers to characterise clinical outcomes (ALT flares and HBV reactivation post HBsAg loss).

Methods: 3 separate cohorts of HBeAg-negative non-cirrhotic patients were studied: *Cohort A:* 66 HBV DNA negative patients on long-term therapy with a nucleoside analogue (NA) *Cohort B:* 25 patients who were long-term suppressed (>5 years) on NA therapy but who stopped NA (follow-up 52 weeks post-cessation); *Cohort C*: 19 patients on long-term NA treatment who achieved HBsAg loss and in whom NA was withdrawn. In serial serum samples from baseline, year 3, year 5 and at time of NA withdrawal and at 52 weeks post-withdrawal HBV DNA (TaqMan [log_{10}IU/ml]), HBsAg (Abbott Architect [log_{10}IU/ml]), HBcrAg (CLEIA, Fujirebio [log_{10}U/ml]), pgHBV RNA (novel real-time PCR research assay from Abbott Diagnostics [LLDQ 1.65 log_{10}U/ml]) were measured. PgHBV RNA detectability was determined in Cohort A patients after 3 years on NA therapy. Rresults in Cohort B were analysed according to severity of post-treatment cessation ALT flares (no flare, mild (2-5xULN ALT) or severe flare (>10ULN ALT). Cohort C patients were divided based on HBV DNA detectability post-cessation.

Results: *Cohort A:* After 3 and 5 years of antiviral therapy 30% and 14% patients still had detectable serum HBcrAg and pgHBV RNA. Patients with detectable pgHBV RNA after 3 years therapy had higher median baseline levels of HBcrAg (4.45 vs.3.15 log_{10}U/ml) and pgHBV RNA levels (2.1 vs.1.79 log_{10}U/ml). C*ohort B:* Post-stopping 9 (36%) patients had no ALT flare, 11 (44%) had mild flares (resolved spontaneously) and 5 (20%) had severe flares (required to restart NA). Detectable HBcrAg and pgHBV DNA at time of NA withdrawal was only observed in patients with a subsequent severe ALT flare. 52 weeks after NA withdrawal 76% patients had detectable HBV DNA, pgHBV RNA was detected in 60% and 80% had HBcrAg detected. *Cohort C*: Only 2 patients had HBV DNA reactivation, but no HBsAg reactivation. Only patients with reactivation had detectable pgHBV RNA, but no HBcrAg and HBV DNA at time of NA withdrawal.

Conclusion: HBcrAg and pgHBV RNA are sensitive biomarkers of continued cccDNA transcription in CHB patients despite marked HBV DNA suppression by NA. These markers were strong predictors of severe ALT flares and HBV DNA reactivation (post HBsAg loss) after NA withdrawal. Their assessment during HBV natural history and on therapy helps to further characterize disease status and management.

OP-04 Hepatitis B core-related antigen correlates with covalently closed circular DNA transcriptional activity in chronic hepatitis B patients

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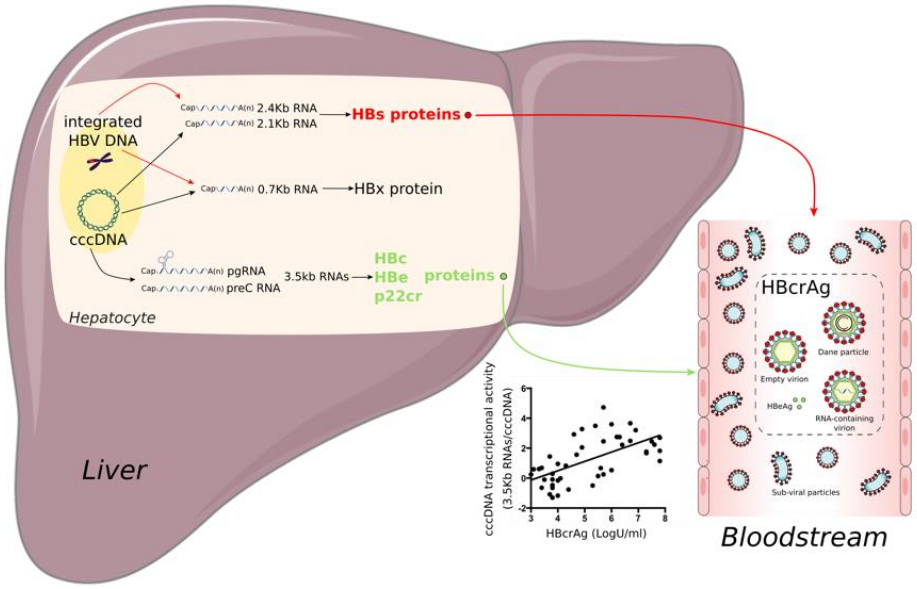
Background and aims: Serum Hepatitis B core-related antigen (HBcrAg) has been proposed to reflect the intrahepatic cccDNA levels, but a comprehensive investigation of its correlation with serum and intrahepatic viral markers and liver histology in a large number of patients is still lacking.
Method: HBcrAg was measured by chemiluminescent enzyme immunoassay (Fujirebio Lumipulse® G HBcrAg) in 130 [36 HBeAg (+) and 94 HBeAg (-)] biopsy proven, untreated, CHB patients. HBcrAg levels were correlated with: a) serum HBV-DNA, quantitative (q)HBsAg and alanine aminotransferase (ALT) levels; b) intrahepatic total (t)HBV-DNA, cccDNA, pgRNA and cccDNA transcriptional activity (defined as pgRNA/cccDNA ratio); c) fibrosis and necroinflammatory activity scores.
Results: HBcrAg levels were significantly higher in HBeAg (+) vs HBeAg (-) patients and correlated with serum HBV-DNA, intrahepatic tHBV-DNA, pgRNA and cccDNA levels and transcriptional activity. Patients who scored negative for HBcrAg (<3 LogU/ml) had less liver cccDNA and lower cccDNA activity as compared to the HBcrAg (+) group. Principal component analysis coupled to unsupervised clustering identified a subgroup of HBeAg (-) patients with higher HBcrAg levels associated to higher serum HBV-DNA, intrahepatic tHBV-DNA, pgRNA, cccDNA transcriptional activity and to higher scores of fibrosis and necro-inflammatory activity.
Conclusion: Our results indicate that HBcrAg is a surrogate marker of both intrahepatic cccDNA and its transcriptional activity that can be useful in the evaluation of new antiviral therapies aiming at a functional cure of HBV infection either by targeting directly or indirectly the intrahepatic cccDNA pool.
Figure:



OP-05 TherVac B-an optimized therapeutic vaccine for hepatitis B

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***TherVac B*-an optimized therapeutic hepatitis B vaccine to cure HBV**

Background and aims: Chronic hepatitis B is characterized by a failure of HBV-specific B and T cell responses while HBV cure is characterized by a loss of HBsAg accompanied by anti-HBs antibody and multispecific CD4 as well as CD8 T cell responses becoming detectable. Therapeutic vaccination is a promising approach to restore insufficient B and T cell immunity in chronic HBV infection. To ensure clinical success, however, improved vaccine design is necessary. We have developed a therapeutic hepatitis B vaccine, termed *TherVac B*, which is based on two protein immunizations using particulate, recombinant HBV S and core antigens (HBsAg, HBcAg) and a boost using a modified vaccinia virus Ankara (MVA) vector expressing HBV core, preS, S and polymerase antigens of different genotypes.
Method: In order to optimize the design of *TherVac B*, we composed and investigated a series of different priming modes including novel HBsAg/HBcAg adjuvant formulations based on either liposomes, or squalene-in-water emulsion (SWE) enriched by saponin QS21 and optionally by monophosphoryl lipid A (MPL), plasmid DNA and RNA priming
Results: Six selected protein antigen formulations proved to be stable and antigens remained intact *in vitro* for at least 2 weeks. *In vivo*, all vaccine formulations were safe and well tolerated. Immunogenicity studies in C57BL/6 mice showed that the new formulations elicited not only very high anti-HBs levels, but also robust HBV-specific CD4 and CD8 T-cell responses outcompeting previously tested adjuvants. In mice, in which persistent HBV replication was established after AAV-HBV infection, immunization with optimized liposomal as well as non-liposomal formulations resulted in a 3-log decrease in serum HBsAg accompanied by very high anti-HBs titers (40, 000-60, 000 mIU/ml), and a pronounced 0.5-1.7-log decrease in serum HBeAg levels. The most vigorous and multifunctional HBV-specific CD4 and CD8 T-cell responses were detected in mice receiving protein antigens formulated with the liposomal adjuvant.
Conclusion: In HBV-transgenic and in AAV-HBV-infected mice, we found that success of therapeutic vaccination largely depends on an appropriate priming, which simultaneously generates neutralizing antibody responses and primes a balanced CD4 as well as CD8 T-cell responses.

OP-06 Investigating the role of HBsAg on cellular immune responses in HBeAg negative patients with chronic hepatitis B to define HBV treatment end points

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Background and aims: The level of hepatitis B surface antigen (HBsAg) produced by HBV-infected hepatocytes could have an immunomodulatory role in HBV infected patients. However, a correlation between HBsAg level and innate and HBV-specific T cell immune responses in HBeAg negative patients with chronic hepatitis B (CHB) has not been well defined. We investigated immune responses in HBeAg negative patients with different levels of HBsAg during the natural course of chronic hepatitis B.
Method: HBeAg negative patients were categorized into 4 groups based on their HBsAg level (HBsAg <100 IU/ml, HBsAg 100-999 IU/ml, HBsAg 1, 000-9, 999 IU/ml, HBsAg ≥10, 000 IU/ml) (n = 44). PBMCs were isolated from these patients and different subsets of immune cells were phenotypically characterized using 14 color flow cytometry. Furthermore, IFNγ+ HBV-specific T cell responses were determined after in vitro culture with overlapping peptides covering polymerase, surface and core of HBV genotype D in the presence or absence of anti-PD-L1 antibody.
Results: Patients with different levels of HBsAg showed similar frequencies of memory, naïve and effector T cells, γδ T cells, Treg cells, MAIT cells, B cells, NK cells, monocytes and DCs. Interestingly, the frequency of γδ T cells, Treg cells and CD1c- myeloid DCs was significantly increased in CHB patients compared to healthy individuals.
Stimulation of PBMCs with HBV overlapping peptides induced core- and polymerase-specific T cell responses. However, surface-specific T cell responses were hardly detectable. We observed a trend towards higher HBV-specific T cells in patients with HBsAg <100 IU/ml. In patients with HBsAg ≥10, 000 IU/ml a high variability of T cell responses were detected. Interestingly, in these patients stronger T cell responses were associated with young age, female gender and low HBV DNA. In general, CD4+ T cell responses were stronger than CD8+ T cell responses. Blocking the PD-1/PD-L1 pathway during in vitro culture significantly increased the core-specific T cell response in patients with HBsAg <100 IU/ml.
Conclusion: Our data suggests that the HBsAg level in HBeAg negative patients per se may have only a minor impact on the innate and cellular immune responses. Patients with HBsAg <100 IU/ml, showed slightly stronger T cell responses to in vitro HBV peptide stimulation especially after using checkpoint inhibitors. Therefore, patients with low HBsAg levels might benefit from immunomodulatory treatment (e.g. checkpoint inhibitors), which might result in functional cure of HBV.

P01-01 In vitro and in vivo models of hepatitis B virus (HBV) replication utilising monomeric HBV genomes to facilitate studies of the HBV cccDNA minichromosome

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Background and aims: The absence of appropriate *in vitro* and *in vivo* replication models enabling direct comparison of hepatitis B virus (HBV) replication across genotypes is hampering the progress of curative HBV research. HBV exists as nine distinct genotypes (A-I) which generally affect particular demographic groups in discrete geographic regions, with varying severity and treatment response. Differences in their replication efficiencies have also been identified and characterized, but molecular analyses have been hindered by the lack of a suitable model that enables thorough investigation of the complete HBV replication cycle, particularly the viral cccDNA reservoir which is a major source of viral persistence and barrier to HBV cure. The Aim of this study was to generate models permitting direct comparison of HBV replication and cccDNA formation in an *in vitro* and *in vivo* setting across HBV genotypes.

Method: Using complete genome PCR, we generated full-length HBV genomes ("1-mers", genotypes A, C and D), transfection of which induced the complete HBV replication cycle, including the generation of a cccDNA-like molecule, *in vitro.* We next introduced these plasmid-free monomeric HBV genomes into mice via hydrodynamic tail-vein injection, to investigate their utility as a novel *in vivo* replication model.

Results: Characterization of HBV replication from liver tissue demonstrated the production of cccDNA-like episomes via Southern blot in both cell culture and murine settings, as well as the detection of major viral RNA transcripts by Northern blot.

Conclusion: Taken together, these *in vitro* and *in vivo* models enable exploration of the interactions between cccDNA-like molecules and viral and host proteins, to further investigate their contributions to HBV replication across genotypes. This may unearth new insights into cccDNA transcriptional regulatory mechanisms and lead to discovery of novel pan-genotypic strategies for the control or inhibition of HBV replication.

P01-02 ICE-HBV and the Global Scientific Strategy for an HBV Cure

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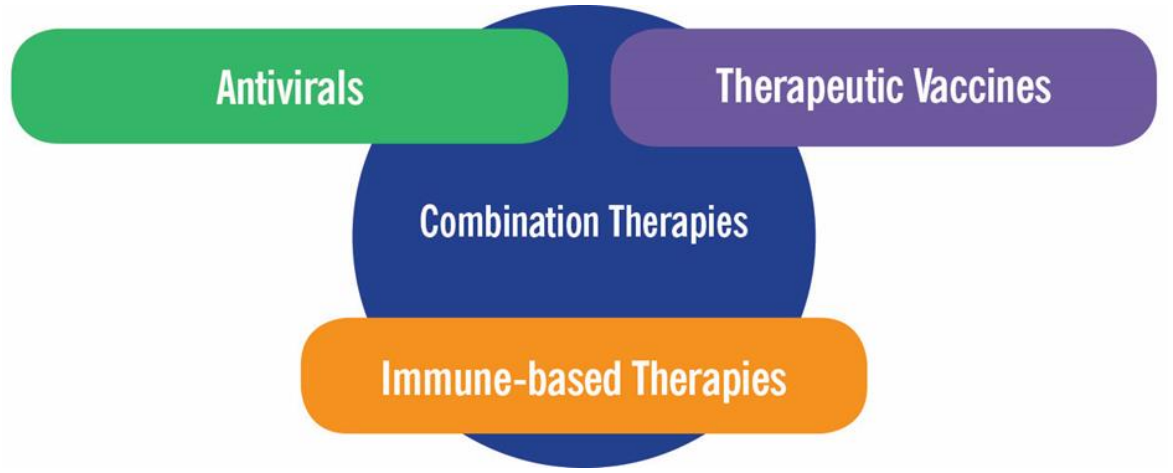
Background and aims: Over 257 million people worldwide are chronically infected with hepatitis B virus (HBV), resulting in over 880, 000 deaths per year from cirrhosis and liver cancer. There is no known cure for chronic HBV, due in part to the continued presence of transcriptionally active DNA in the nucleus which is not directly targeted by current antiviral therapies. Our aim is to inspire and support the discovery of a safe, scalable and effective cure for the benefit of all people living with CHB.
Method: To achieve this, the International Coalition to Eliminate Hepatitis B (ICE-HBV) is coordinating collaborative partnerships among researchers and stakeholders to accelerate the search for an HBV cure.
Results: ICE-HBV working groups have developed a joint global scientific strategy for HBV cure, with input from key stakeholders including the HBV affected community. Through engagement with key stakeholders, ICE-HBV aims to drive changes in governmental policy to ensure more funds are channelled to HBV cure research and drug development. ICE-HBV fosters new collaborations among HBV researchers and industry worldwide and initiate new projects to fast-track the discovery of an HBV cure such as animal models for HBV, serum biomarkers, point-of-care assays, cost-effectiveness modelling, community engagement and scientific literacy, health policy.
Conclusion: The push for a cure for chronic HBV infection is particularly timely thanks to the recent development of cell culture infection models that, for the first time, empower truly curative research. Through its global network, ICE-HBV is striving to promote effective collaboration and facilitate opportunities that can produce a cure for chronic HBV infection.
Figure: Therapeutic Strategies



P01-03YI HBV RNA at 6 months predicts HBeAg loss in nucleos (t)ide analogue treated HBeAg positive patients: demonstration of clinical utility?

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Background and aims: HBeAg loss in patients with HBeAg-positive chronic hepatitis B is an important treatment end point and a pre-requisite for functional cure. Only around half of patients achieve HBeAg loss after five years of treatment with nucleos (t)ide analogues (NUC), despite nearly all achieving profound HBV DNA suppression. It is still unclear, but critical to understand, whether newer biomarkers differentiate patients who will achieve serological end points such as HBeAg loss. In this single-centre retrospective longitudinal cohort study, we evaluated HBV RNA and hepatitis B core-related antigen (HBcrAg) concentrations in NUC treated HBeAg-positive patients, with the aim of characterising differences that predict HBeAg loss.

Method: Consecutive HBeAg-positive patients started on NUC therapy in 2012 with adequate stored serum samples were analysed. Baseline characteristics included age, sex, HBV genotype, ALT and liver fibrosis assessment by transient elastography or histology. Serum from baseline, 3, 6 and 12 months into NUC therapy and last follow-up were tested for HBV DNA, quantitative hepatitis B surface antigen (HBsAg), HBcrAg and HBV RNA concentrations.

Results: 28 patients received treatment with tenofovir disoproxil fumarate. After median follow-up of 64 months, 17 patients (61%) achieved HBeAg loss and 26 patients (93%) were HBV DNA negative. There were no significant differences at baseline between patients who subsequently lost HBeAg and those who remained positive. There was a strong correlation at baseline between HBV DNA and HBcrAg ($r = 0.862$, $p < 0.001$), and HBV DNA and RNA ($r = 0.704$, $p < 0.001$). This correlation was subsequently lost with treatment.

During treatment, median HBV DNA and quantitative HBsAg were similar between the groups at all time points (Figure 1). In contrast, early differences in HBcrAg concentrations were seen at 3 months ($p = 0.019$) and HBV RNA at 6 months ($p = 0.006$); by last follow-up, patients who achieved HBeAg loss had significantly lower HBcrAg and HBV RNA levels (median HBcrAg 5.8 vs 3.8 logU/ml, $p = 0.026$; median HBV RNA 3.94 vs. 0.83 logU/ml, $p < 0.001$). HBV RNA decline from baseline at 6 months was greater in those who achieved HBeAg loss ($p = 0.038$). Furthermore, HBV RNA decline had good predictive value for HBeAg loss; a 1 logU/ml decline at 6 months had a positive predictive value of 71.4% and area under the receiver operating curve of 0.747.

Conclusion: In this study, HBV RNA decline after 6 months of NUC therapy predicted HBeAg loss with good performance. HBV RNA decline presents an important goal for novel therapies, with HBV RNA suppression potentially being associated with favourable treatment end points.

Figure:

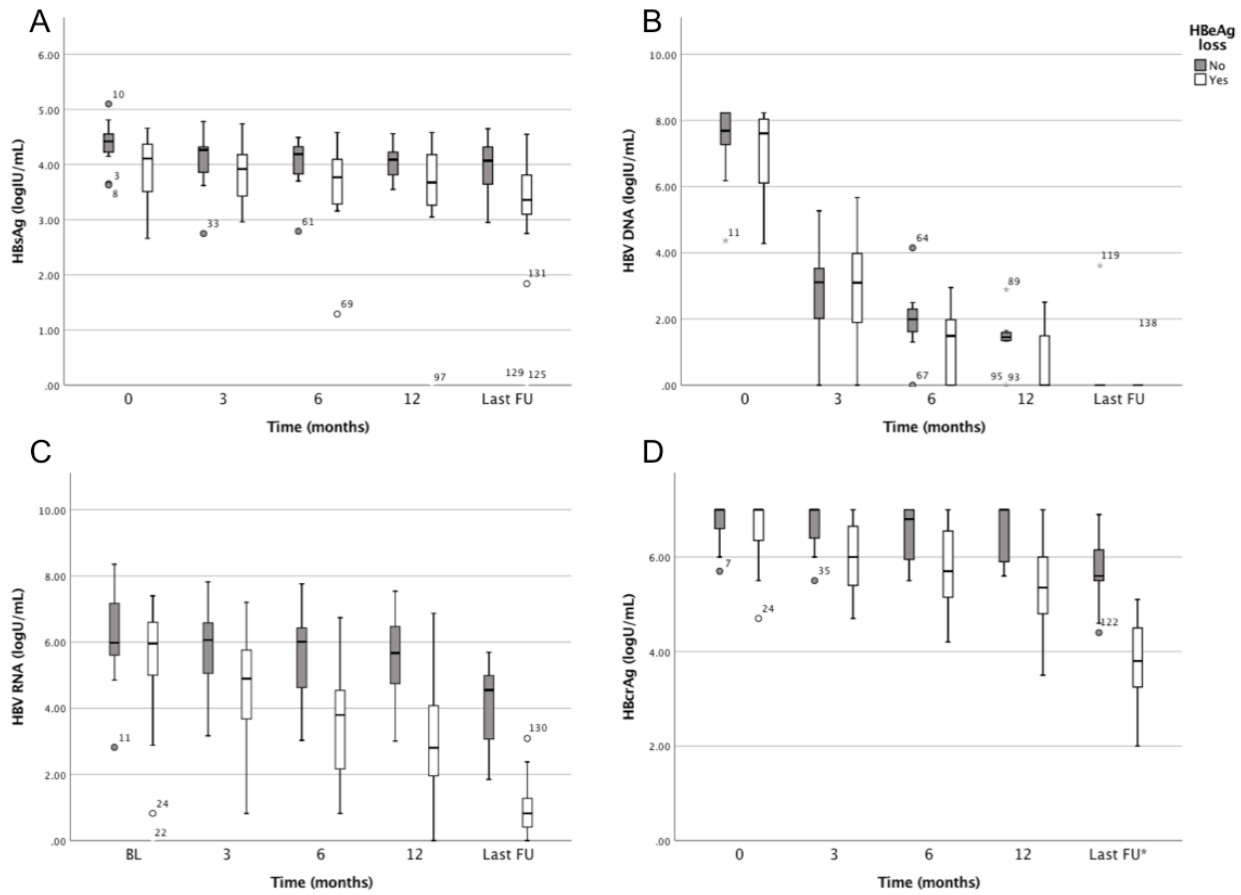

Figure 1. Box plots of HBsAg (A), HBV DNA (B), HBV RNA (C) and HBcrAg (D) concentrations during treatment with nucleos (t)ide analogues in HBeAg-positive patients (*last FU; median duration 64 months).

P01-04 Dynamic changes of serum HBV pgRNA levels in patients with chronic hepatitis B treated with entecavir or peg-interferon

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Background and aims: Serum hepatitis B virus (HBV) pregenome RNA (pgRNA) levels may independently predict virological and serological response during antiviral therapy. This study aimed to find the correlation between serum HBV pgRNA levels and other biomarkers, further investigate the dynamic changes of serum HBV pgRNA levels and its clinical significance during treatment.

Method: A real-time polymerase chain reaction (PCR) was developed for quantitative analysis. HBV pgRNA levels were retrospectively determined in serial serum samples from 136 patients with chronic HBV infection who have received entecavir or peg-interferon treatment. Receiver operating characteristics (ROC) analysis was performed to evaluate the prediction value in HBeAg seroconversion of individual biomarkers.

Results: The mean serum HBV pgRNA level was 6.41 [1.94] copies/ml at baseline, which was higher in HBeAg-positive patients of 7.20 [1.54] copies/ml than in HBeAg-negative patients of 4.60 [1.51] copies/ml ($p < 0.001$). Baseline HBV pgRNA levels correlate strongly with HBV DNA levels ($r = 0.82$, $p < 0.001$), moderately with HBsAg levels ($r = 0.69$, $p < 0.001$), and weakly with serum alanine aminotransferase ($r = 0.28$, $p < 0.05$). Peg-IFN treatment induced a stronger decline in HBV pgRNA level from baseline to week 4, 24 and 48 in comparison to entecavir (ETV) monotherapy ($p < 0.05$). ETV treated patients with HBeAg seroconversion showed a stronger decline in HBV pgRNA level at week 4, 12, 24 and 48. At baseline, the area under ROC (AUC) of HBV pgRNA was 0.68, comparable to those of HBV DNA (AUC = 0.66) and HBsAg (AUC = 0.64) in ETV treated patients; however, in peg-IFN treated parents, the AUC of HBV DNA (AUC = 0.66) was highest in predicting seroconversion, followed by HBV pgRNA and HBsAg (AUC = 0.63 and 0.60). No significant difference was noted for AUCs among these biomarkers ($p > 0.05$). During treatment, the best prediction of HBeAg seroconversion in ETV treated patients was HBV pgRNA at week 4 (AUC = 0.71, the corresponding cut-off value, sensitivity, specificity, PPV and NPV were 7.95 log10copies/ml, 83%, 56%, 40%, 91%), while in peg-IFN treated patients, HBV pgRNA level at week 24 allowed the best prediction (AUC = 0.70, the corresponding cut-off value, sensitivity, specificity, PPV and NPV were 3.55 log10copies/ml, 89%, 62%, 44%, 94%).

Conclusion: Serum HBV pgRNA levels may serve as a brand new biomarker in the evaluation of patients with chronic HBV infection during antiviral therapy.

P01-05 Adverse events of nucleos (t)ide analogues for chronic hepatitis B: a systematic review

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Background and aims: The nucleos (t)ide analogues (NAs) are the main drugs used in chronic hepatitis B treatment, leading to undetectable HBV DNA levels in the vast majority of patients. A significant number of patients have been treated with NAs, increased the experience with the efficacy, resistance and safety profile over the years. Despite the fact that NAs have a favorable safety profile, undesired adverse events (AEs) may occur during the treatment. Given the eminent number of patients receiving NAs worldwide, even a small risk of any of these toxicities can be translated into a major medical issue. The main objective of this systematic review is to analyze information on AEs described as related to the use of NAs in published studies.

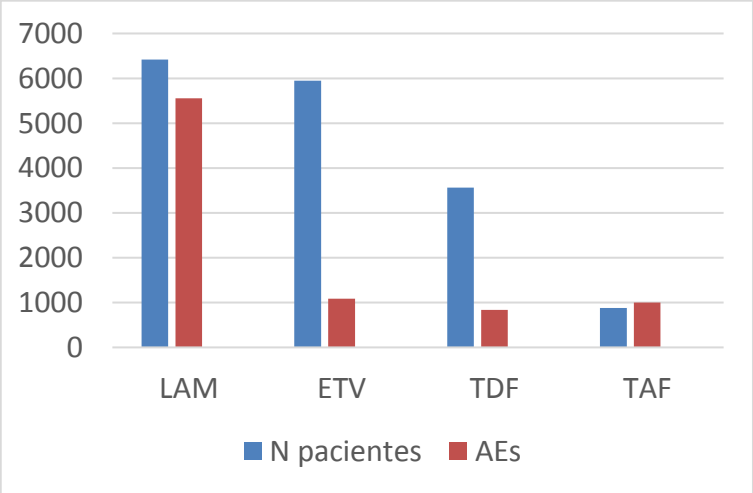
Method: We chose the following MesH terms: chronic hepatitis B, side effects and treatment. All articles published from 1 January 1990 up to 19 February 2018 in MEDLINE of PubMed, EMBASE, the Cochrane Library and LILACS databases were searched. We included studies published in English language. We followed an established protocol which had been registered in PROSPERO (International prospective register of systematic review), and the record is available on https://www.crd.york.ac.uk/PROSPERO/ (Registration number: CRD42018086471)

Results: A total of 108 articles were selected for analysis. There were 6419 patients treated with lamivudine (LAM), 5947 treated with entecavir (ETV), 3566 treated with tenofovir disoproxil fumarate (TDF), and 876 treated with tenofovir alafenamide (TAF). The most common AEs with all drugs are abdominal pain/discomfort, nasopharyngitis/upper respiratory tract infections, fatigue, and headache.

TAF was the drug with the lowest number of patients and the highest number of adverse events (AEs) reported per patient (1.14 AE/treated patient). When TDF was compared to TAF, the reduction of bone density was greater with the use of TDF, although no drug-related fractures were described. The same occurred with the glomerular filtration rate, also with a greater reduction in the groups that received TDF. However, the curious aspect was that renal/urinary changes were the 4th group with the highest reports of AEs with TAF. When analyzing these AEs, there were 43 cases of glycosuria (versus 2 cases with TDF) and 68 cases of urine erythrocytes (versus 24 with TDF).

Conclusion: Treatment of CHB with NAs is safe, with a low incidence of adverse events. The most common AEs with all drugs are abdominal pain/discomfort, nasopharyngitis/upper respiratory tract infections, fatigue, and headache. TDF demonstrated a greater reduction in the glomerular filtration rate and bone density of the lumbar spine and hips when compared to TAF. Currently, the number of patients treated with TAF is still too small to consolidate that TAF is really safer when compared to TDF.

Figure: Number of patients treated and absolute number of adverse events reported for each drug



P02-01 Factors associated with hepatitis -B virus infection among students in Krachi senior high school in the Krachi west district of the Volta region

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Background and aims: Hepatitis B virus (HBV) occurs globally and is of a great public health importance. According to World Health Organization (WHO), “more than two (2) billion people worldwide have been infected with Hepatitis-B virus and about 350 million people remain chronically infected”. Even though Ghana belongs to one of the areas where HBV is highly endemic not much study detailing the burden of the disease among the youth has been conducted. This work was intended to determine the burden of HBV infection and its associated risk factors among students in the Krachi Senior High school in the Krachi West district of the Volta region.
Method: Using the 2017 student register as sample frame, 182 students were randomly selected for this cross-sectional sero prevalent survey. Chi-square test and Student t- test were used to compare categorical and continuous variables. Those categorical variables with p <0.200 were analyzed in a final regression model.
Results: The sero- prevalence of HBV among the 182 study participants was found to be 14.3%. More females than males 14/26 (53.8%) tested positive for the HBsAg. The highest prevalence was recorded in 18-20 age groups 14/87 (16.09%). Place of barbering was found to be statistically significant with HBV **(aOR**: 0.48 95%CI.0.28-0.84) (p = 0.01).Also sharing of same spoon/cutlery among students also significantly influences HBV infection among students **(aOR:** 0.46, 95%CI.0.26-0.8) (p = 0.01)
Conclusion: Hepatitis B virus infection is significantly high among students attending Krachi West Senior High School. Place of barbering and sharing of cutlery among students are factors significantly associated with the virus infection

P02-02YI Long-term safety and efficacy of tenofovir disoproxil fumarate (TDF) in nucleos (t)ide (NA) analog-experienced chronic hepatitis B patients

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Background and aims: One major challenge in the treatment of chronic hepatitis B (CHB) is to maintain long-term viral suppression without promoting the selection of drug-resistant mutations and drug related toxicity. Tenofovir disoproxil fumarate (TDF) has demonstrated high antiviral efficacy in treatment-naïve patients with chronic HBV infection but experience in nucleoside/nucleotide analogue (NA)-experienced patients is limited. This study assessed the efficacy and safety of TDF up to 10 years of NA-experienced CHB patients.
Method: We retrospectively analyzed this prospectively maintained data of 132 TDF treated patients from 2006 to 2010 at tertiary care centre. For each patient, medical records were reviewed to obtain demographic information, baseline CHB characteristics, and co morbidities. Multivariate logistic regression analyses were carried out to identify the unique association between predictor variables—including age, sex, diabetes, hypertension, pre-existing renal insufficiency, history of renal transplant, duration of therapy—and increase in SCr (serum creatinine) levels or decrease in eGFR. For all analyses, a *P* value of <0.05 was considered to be statistically significant.
Results: Patients demographics included 121 males and 11 females with mean age of 54 years. Out of 132 patients, 100 had chronic hepatitis and 32 had cirrhosis. Before the treatment had been started, mean HBV DNA, alanine aminotransferase (ALT) levels were 5.18 Log $_{10}$ IU/ml, 128 mU/ml respectively. After mean duration of 6.3 years of TDF therapy 125 (94.6%), 123 (93%), 67 (94.3%) patients achieved a virological response, ALT normalization and HBe antigen seroconversion respectively.7 patients did not come for follow-up. 3 patients developed HBs antigen clearance and out of them, 2 developed HBs antigen seroconversion.4 patients developed HCC, out of them, 3 were expired.17 (12.8%) patients developed renal dysfunction defined by eGFR <50ml/min by Cockcroft-Gault formula. By multivariate analysis, history of diabetes (p = 0.003) and pre-existing renal insufficiency (p = 0.001) were the only significant factors associated with an increase in SCr levels (≥0.2mg/dL).
Conclusion: TDF is effective and safe for NA-experienced CHB patients and should be used cautiously in patients with co-morbidities like diabetes and pre existing-renal insufficiency, due to risk of renal dysfunction.

Table: Factors Associated with Increased SCr levels (≥0.2 mg/dL) during Treatment

Factor	Adjusted OR	95% CI	*P* value
Age	1.000	0.933-1.072	.998
Sex	0.95	0.001-6.517	.276
History of diabetes	0.057	0.008-0.383	**.003**
Hypertension	0.660	0.099-4.420	.669
Pre existing-renal Insufficiency	0.010	0.001-0.145	**.001**
Duration of therapy	1.122	0.767-1.642	.553
History of renal transplant	0.090	0.003-3.049	.181

P02-03YI The gamma glutamyl transpeptidase to platelets ratio and gamma glutamyl transpeptidase to albumin ratio do not correlate to fibroscan measurement of hepatic fibrosis in chronic hepatitis b egyptian patients: a pilot study

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Background and aims: The gamma-glutamyl transpeptidase to platelet ratio (GPR) and the gamma-glutamyl transpeptidase to albumin (GAR) are novel biomarkers suggested to assess fibrosis. Being routine cheap and widely available it was suggested to be used in resource limited countries for prognosis and follow-up in chronic infection with hepatitis B virus (HBV). The aim of this work was to assess the correlation between these markers (GPR and GAR) and degree of fibrosis and steatosis in HBV patients as measured by fibroscan.

Method: After ethical approval of the protocol, 30 chronic HBV patients were recruited from tropical medicine department -Tanta university. They underwent fibroscan examination including transient elastogram for fibrosis and Capture attenuated parameter (CAP) for steatosis with concomitant testing of liver functions and complete blood picture. The GPR and GAR ratios were calculated. The relation between GPR and GAR ratios to fibrosis and steatosis were tested using Pearson rank correlation.

Results: There was no significant correlation between GPR and GAR ratios and fibrosis ($p = 0.291$ and 0.148) or steatosis ($p = 0.522$ and 0.271). on the other hand, GPR had significant positive correlation to GAR ($p < 0.0001$). also, fibrosis was positively correlated to steatosis ($p = 0.012$) and ALT ($p = 0.03$).

Conclusion: Despite being cheap and non-invasive, GAR and GPR were not correlated to fibrosis or steatosis in our patients. We cannot recommend their use as surrogate markers for fibrosis in HBV patients.

P02-04 Core outcome set for Cochrane reviews on chronic hepatitis B infection

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Background and aims: Pre-specification and definition of outcomes prevents data-driven analyses. The use of agreed core outcome sets in systematic reviews of interventions benefits trial investigators and systematic review authors in providing useful synthesis of data. Clinically relevant and patient-centred outcome measurements assist physicians in their selection of the best-available evidence-based choice of treatment. Therefore, we aimed at developing core outcomes for chronic hepatitis B virus (HBV) infection.
Method: Cochrane Hepato-Biliary Group editors together with review authors have prespecified the outcomes for chronic HBV infection and their definitions. We achieved this through face-to-face meetings, telephone discussions, and correspondence. During our work, we used http://www.comet-initiative.org; The International Conference on Harmonization (ICH) harmonised tripartite guideline. Guideline for good clinical practice CFR and ICH Guidelines. Vol. 1. Pennsylvania, USA: Barnett International/PAREXEL, 1997; PLoS Med 2017; 14: e1002447; and https://www.cochranelibrary.com. We are in the process of involving patient representatives.
Results: We advise the use of:
Primary outcomes: a) proportion of trial participants with all-cause mortality; b) health-related quality of life (validated questionnaires); c) proportion with serious adverse events according to ICH Guidelines for Good Clinical Practice 1997.
Secondary outcomes: a) HBV-related morbidity (i.e. proportion of participants who developed cirrhosis, ascites, variceal bleeding, hepatorenal syndrome, hepatocellular carcinoma, hepatic encephalopathy, or underwent liver transplantation); b) proportion with non-serious adverse events (i.e. any adverse event that do not meet the above criteria for serious adverse events).
Exploratory outcomes: proportions a) with specific serious adverse events separately analysed; b) with non-serious adverse event separately analysed; c) without histological improvement; d) without HBsAg to anti-HBs seroconversion; e) with detectable HBV DNA in serum or plasma; f) with detectable HBeAg and without HBeAg seroconversion (only relevant for HBeAg-positives).
Conclusion: Pre-specification and definition of patient-centred clinical outcomes is recommended for systematic reviews, which should also impact design and reporting of primary trials.

P02-05YI Outcomes of severe acute viral hepatitis b in Latvia

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Background and aims: Approximately two billion people worldwide have evidence of past or present infection of hepatitis B. In acute viral hepatitis B successful clearance of virus is more than 95% of immunocompetent patients. Only 5% of patients develop chronic hepatitis B virus infection. Chronic infection is a major global health problem already and puts people at high risk of death from cirrhosis and liver cancer. Fulminant hepatitis occurs rarely-0.1-0.5%. The aim of this study was to analyze outcomes of severe acute viral hepatitis B in Latvia.
Method: This retrospective study included patients (n = 40) with severe acute viral hepatitis B (AVH-B). All patients were treated in Infectology Center of Latvia from January 2005 to December 2017. Patients were classified as having severe AVH-B if they fulfilled any 2 of 3 criteria: hepatic encephalopathy, serum bilirubin ≥10.0 mg/dL and international normalized ratio (INR) ≥1.6. Statistical analysis was done, using SPSS version 20.0
Results: Study included 40 patients with mean age 47 ± 16 years and average duration of hospitalization 21 days. 19 (46.9%) were males and 21 (53.1%) females. All patients (n = 40) had severe acute viral hepatitis B treated with Lamivudine 100 mg daily. Three criteria of severe hepatitis was found in 19 (59%) of cases. None of the patients developed chronic hepatitis. In 23 (57.5%) cases serum HBsAg and HBeAg disappeared and they recovered. Ten patients (25%) had unknown outcome, because they lost from follow-up, but 7 (17.5%) patients died. Statistically significant more severe coagulopathy was in patients group with lethal outcome, in comparison to patients in recovery group, INR = 3.10 vs. INR = 1.83, respectively, p = 0.046. However, statistically significant difference between bilirubin, alanine aminotransferase and aspartate aminotransferase levels was not observed. All patients of lethal group had all 3 severe hepatitis criteria. 7 patients had fulminant hepatitis which resulted with liver coma and death. Two patients of lethal group had co-infection: chronic viral hepatitis C and acute viral hepatitis D. Three patients were immunosuppressed: one of them had prostate cancer, one had Hodgkin's lymphoma, one patient had tetraparesis after vertebral fracture. One patient had Wilsons disease with cirrhosis.
Conclusion: Immunosuppressed patients or patients co-infected with other liver diseases more frequently had fulminant hepatitis or acute liver failure, which resulted in liver coma. Patients with fulminant or severe hepatitis must be considered for liver transplantation, because these patients have higher risk of acute liver failure. Method of treatment mainly is liver transplantation, unfortunately, it is available in Latvia only since 2018.

P03-01 Evaluation of the Safety and Tolerability of Transaminase Flares During Antiviral Therapy in Patients with HBeAg Negative Chronic HBV Infection or HBV/HDV Co-infection

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Background and aims: The onset of transaminase flares during antiviral therapy of chronic HBV infection or HBV/HDV co-infection is an important consideration in the pursuit of functional cure of HBV and HDV infection. The high proportion of transaminase flares observed during NAP-based therapy in HBV infection (REP 401 study NCT02565719) or HBV/HDV co-infection (REP 301 study NCT02233075) affords a unique opportunity to evaluate factors predicting transaminase flares during therapy, the tolerability of different flare geometries during therapy and how flares correlate with antiviral response during therapy and the establishment of functional

Method: Data from all 52 participants from the REP 301 (12) and REP 401 (40) studies were included in the analysis. Baseline data used in the analysis was HBsAg, ALT, AST, GGT and median hepatic stiffness (MHS, as measured by Fibroscan). Serial on-therapy biochemistry data included ALT, AST, GGT, Alk. Phos, bilirubin, albumin, INR and MHS at the end of treatment and during follow-up. Serial virologic data included HBsAg, anti-HBs, HBV DNA and HDV RNA.

Results: Three different transaminase flare geometries were observed during therapy: a single flare self-resolving during therapy, a single flare with a persistently elevated transaminase tail during therapy and a “double” flare with two distinct peaks.

Flare geometries, transaminase maxima or transaminase AUC during therapy were not correlated with baseline HBsAg, ALT, AST, GGT or MHS (up to 30.7 kPa in this participant population). No alterations in bilirubin, albumin or INR were observed with any flares and patients were asymptomatic throughout these flares including the absence of jaundice. Increased MHS at end of therapy was asymptomatic and correlated with transaminase AUC but decreased or normalized during follow-up.

The occurrence of transaminase flares was correlated with the introduction of pegIFN in patients who experienced HBsAg reduction and was more pronounced in patients where HBsAg reduction became <1 IU/ml. Flares were also accompanied by pronounced increases in circulating anti-HBs during therapy. The strength of transaminase flares was also correlated with the establishment of functional cure of HBV or HDV or the establishment of inactive chronic HBV.

Conclusion: Transaminase flares are very common during NAP-based therapy of HBV infection or HBV/HDV co-infection and appear well tolerated, suggesting that in the context of these infections, transaminase flares can occur without impacting liver function. Flares are also correlated with HBsAg reduction, increases in circulating anti-HBs and the establishment of functional control/cure of HBV and HDV infection, suggesting they are a component of a restored immunological control of these infections.

P03-02 Efficient Clinical Development of More Curative HBV Therapies

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Background and aims: HBV causes 600, 000-1 million deaths/yr (1 death every 1-2 min). Registration trials for HBV nucleos (t)ide (nuc) antivirals consumed 6-9 years to initial regulatory filings, with 1-5+ yrs for global regulatory approvals. Efficient development of more curative therapies could avert several million HBV-related deaths

Method: With current HBV therapies durable post-treatment responses are associated with lowered HBsAg levels- (e.g. sAg <<1000 IU/ml), which putatively relieve sAg-related `decoy' suppression of HBV immune responses. Initial trials of new HBV therapies should include untreated pts (the global majority) and nuc-suppressed pts. To establish an early efficacy profile justifying Phase 2-3 trials, Phase 1b treatment (Rx) with new agents should be up to 12 wks, to assess serum HBV DNA and HBV RNA declines, quantitative sAg and eAg, and any other markers needed to confirm efficacy mechanisms for novel agent (s). Early Phase 2 trials in untreated HBV pts should be controlled studies of *de novo* combination Rx (comboRx) with investigational agent (s) plus an HBV nuc vs. placebo-blinded nuc monoRx. Additive or synergistic efficacy can be assessed by comparisons of 1st- and 2nd-phase HBV DNA decline kinetics and changes in sAg and eAg levels. With potent nucs 1st-phase HBV DNA decline is rapid and profound; superior efficacy for comboRx is likely to be more evident in improved 2nd-phase HBV DNA declines. `Functional cure' to the point of non-detectable sAg is a worthy goal, but as a 1° end point it may unnecessarily prolong Phase 2-3 trials, with delays in product registrations. Many pts have appreciable serum sAg levels putatively derived from integrated HBV DNA sequences, not cccDNA. As low-level viremia has residual HCC risks, it is desirable to treat patients to a point of non-detectable HBV DNA and lowered sAg levels (to <<1, 000 IU/ml). Rather than treat to sAg seronegativity, however, Phase 2-3 trials could include testing of pt sera with the HBV core-related antigen (HBcrAg) assay, which detects 3 HBV core-related peptides reflecting transcriptional activity of cccDNA. In pts who achieve non-detectable HBV DNA, with lowered sAg levels and negative serum HBcrAg, residual serum sAg may be derived from integrated sAg sequences. Although they may still be sAg-seropositive, evaluation of treatment discontinuation would be reasonable for such pts, with follow-up for response durability.

Conclusions:

Trials of new HBV agents in untreated pts will recruit more rapidly if all randomized treatment assignments include at least one proven HBV antiviral (nuc or peg-IFN). Efficacy can be assessed through 1st- and 2nd-phase viral kinetics, and Rx discontinuation can be tested when HBV DNA is non-detectable, sAg levels are low, and negative HBcrAg assays suggest negligible transcription from pts' hepatic cccDNA

P03-03 Prediction of HBeAg loss on long-term nucleos (t)ide analogues with a novel HBV pgRNA assay

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Background and aims: With prolonged nucleos (t)ide analogues (NAs) therapy, only 40-50% achieve HBeAg loss. We aim to identify the virological profiles of those unlikely to clear HBeAg so alternative novel therapy can be considered.

Method: HBeAg (+) patients with baseline HBV DNA ≥5 log_{10} IU/ml on NAs for ≥20 months with stored sera were included. Clinical parameters were compared between patients with and without HBeAg clearance. All patients continued therapy after HBeAg loss. HBV RNA by Abbott research assay (Sensitivity = 1.65 log_{10} U/ml; linearity 2.5-7.5 log_{10} U/ml). Quantitative HBsAg by Abbott ARCHITECT.

Results: This predominantly Asian (86%) cohort included 17 and 11 patients with and without NA treatment-related HBeAg loss respectively. Their baseline clinical parameters were similar [Table]. Both groups achieved HBV DNA <20 IU/ml after similar treatment duration. Those with HBeAg loss had significantly greater HBV RNA decline when HBV DNA became <20 IU/ml. The decline of qHBsAg was similar in the 2 groups when DNA was suppressed. Six of 17 (35%) with HBeAg loss had ALT >100 U/L associated with 1.7 log (mean) HBV RNA reduction preceding HBeAg clearance. Their HBV RNA levels became <1.65 log_{10} U/ml after HBeAg loss. In contrast, 11 of 17 patients without hepatitis flare had higher HBV RNA (Mean 3.1 log_{10} U/ml) at time of HBeAg clearance. With long-term therapy, HBV RNA levels remained >4 log_{10} U/ml among those who were still HBeAg (+). In contrast, 10 of 17 (59%) with HBeAg clearance achieved HBV RNA <1.65 log_{10} U/ml. At last follow-up, qHBsAg was lower for those with HBeAg loss but did not reach significance. One patient achieved undetectable HBV RNA and HBsAg four months after HBeAg loss.

Conclusion: On treatment HBV RNA kinetics may differentiate patients with and without HBeAg clearance on long-term NAs. Patients with >3 log decline in HBV RNA when HBV DNA was suppressed (<20 IU/ml) were more likely to achieve HBeAg loss. If their HBV RNA remained >4 log_{10} U/ml with continuous therapy, the chance of losing HBeAg was unlikely. Hepatitis flare was often associated with significant HBV RNA reduction and subsequent HBeAg clearance. These observations need to be carefully validated.

Table:

	HBeAg Loss (N = 17)	**Without HBeAg Loss (N = 10)**	**P Value**
Duration of therapy: Months	75 (22, 168)	84 (27, 184)	0.3
Baseline HBV DNA (log_{10} IU/ml)	7.2 (5.2, 8.2)	7.5 (5, 8.2)	0.28
Baseline ALT (U/L)	107 (18, 260)	84 (14, 268)	0.2
Time to HBV DNA <20 IU/ml (Months)	38.6 (10.6, 141)	46.9 (6, 159)	0.29
qHBsAg (log_{10} IU/ml) when HBV DNA <20 IU/ml	3.1 (0, 4.8)	3.6 (3, 4)	0.09
qHBsAg decline from baseline when HBV DNA <20IU/ml	0.8 (0, 3.3)	0.7 (0, 1.4)	0.4
HBV RNA (log_{10} U/ml) when HBV DNA <20 IU/ml	2.5 (*UD, 5.3)	5.6 (3.8, 6.7)	**<0.00002**
HBV RNA decline from baseline when HBV DNA <20IU/ml	3.3 (0.3, 6.3)	1.3 (0.2, 3.2)	**0.001**
HBV RNA at last follow-up	1.6 (*UD, 3.3)	4.7 (3.3, 5.8)	**<0.00001**
qHBsAg at last follow-up	2.8 (*UD, 4.5)	3.3 (2.9, 3.9)	0.07

*UD = undetectable

P03-04YI Diagnostic accuracy of aspartate transaminase to platelet ratio index, fibrosis index based on 4 factors (fib-4) and golgi protein 73 for prediction of significant liver fibrosis in chronic hepatitis b patients:an egyptian cross-sectional study

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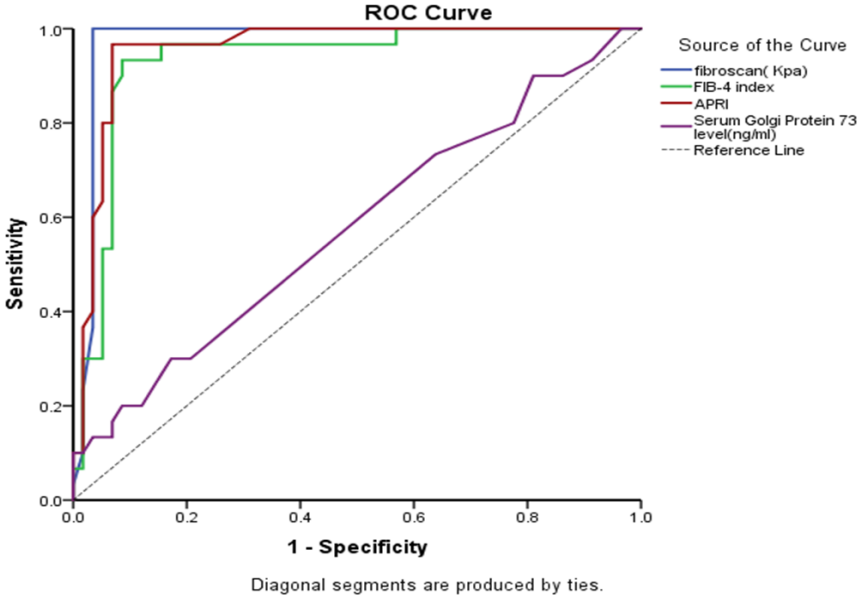
Background and aims: Evaluation of liver fibrosis is an essential clinical tool that guides the therapeutic decision and helps in predicting the prognosis for patients with chronic hepatitis B virus (HBV) infection.The aim of this study is to evaluate the accuracy of aspartate transaminase-to-platelet ratio index (APRI) and fibrosis index based on 4 factors (FIB-4)-two easily calculated non-invasive scores-and serum Golgi protein 73 (GP73) to predict significant liver fibrosis in chronic hepatitis B patients compared to transient elastography (TE).

Method: This is a cross sectional study conducted on 88 antiviral-naïve chronic HBV patients attending to Cairo University center for hepatic fibrosis over the period between January 2018 and September 2018. Enrolled patients had undergone thorough clinical examination, assessment of biochemical parameters and abdominal ultrasound . Serum GP73 was quantified using enzyme-linked immunosorbent assay (ELISA) kits. Liver stiffness measurement was performed using transient elastography .APRI and FIB-4 scores were calculated using the following formulas: FIB-4 = age (years) × AST/platelet count × (ALT) 1/2), APRI = (AST/upper limit of AST)/platelet count × 100.The receiver operating characteristic (ROC) curves were used to analyze the diagnostic accuracy of the studied serologic markers for prediction of significant hepatic fibrosis .

Results: Out of 88 patients who were enrolled, 66 % were males. The mean age was 40 ± 10 years. Thirty-four % of patients had significant liver fibrosis as evident by TE. Both APRI and FIB-4 showed good diagnostic performance in prediction of significant fibrosis (p value <0.001); APRI had area under receiver operating characteristic (AUROC) curve of 0.957 (95% CI: 0.914-0.999) with best cut off 0.575 at which sensitivity and specificity were 96.7% and 93.1% respectively. FIB-4 showed AUROC of 0.931 (95% CI: 0.873-0.99) with best cut off 1.35 at which sensitivityand specificity were 96.7% and 84.5% respectively.Serum GP73 provided poor diagnostic value for prediction of significant liver fibrosis (p value 0.233) and AUROC was 0.578 (95% CI: 0.450-0.706).

Conclusion: APRI and FIB-4 showed comparable diagnostic accuracy to TE and are reliable tools for the diagnosis of significant hepatic fibrosis in antiviral-naïve chronic hepatitis B patients .However, serum GP73 couldn't predict significant liver fibrosis but this findings need further assessment on a large scale of patients.

Figure:



P03-05YI Serum sulfatase-2 is a new prognostic marker of patients with hepatitis B Virus-related hepatocellular carcinoma

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Background and aims: Sulfatase-2 (SULF-2), an extracellular enzyme promoting tumor proliferation, has been shown to be upregulated in liver tissues of patients with hepatocellular carcinoma (HCC). However, the diagnostic and prognostic roles of this marker in sera of patients with HCC are currently unknown. This study was aimed at investigating the clinical usefulness of serum SULF-2 in Thai patients with hepatitis B virus (HBV)-related HCC.
Method: Three studied groups included 146 patients with HCC, 119 patients with non-malignant chronic HBV infection (non-HCC) and 50 healthy subjects. Serum SULF-2 and alpha-fetoprotein (AFP) levels were measured by enzyme-linked immunosorbent assay (ELISA) method.
Results: The HCC group was significantly older with higher proportion of male compared with the non-HCC group and healthy controls ($p < 0.001$). Patients with HCC had significantly higher levels of serum SULF-2 compared to patients without HCC and controls (27.3 ± 10.3 vs. 18.5 ± 5.2 vs. 15.8 ± 4.3 ng/ml, $p < 0.001$). The area under the curve (AUROC) for differentiating HCC from the other groups were 0.79 (95%CI; 0.73-0.84, $p < 0.001$) for SULF-2 and 0.90 (95%CI; 0.86-0.94, $p < 0.001$) for AFP. In the HCC group, serum SULF-2 levels positively correlated with AFP levels ($r = 0.461$, $P = 0.001$), Child-Pugh classification ($r = 0.206$, $P = 0.016$), tumor size ($r = 0.277$, $P = 0.001$) and tumor stage (BCLC stage) ($r = 0.274$, $P = 0.001$). High SULF-2 level (above median value as a cut-off point of 20 ng/ml) was significantly correlated with poor overall survival and was an independent prognostic factor in patients with HCC.
Conclusion: Elevated serum SULF-2 levels were associated with advanced tumor size and stage, suggesting that this novel marker might play important roles in promoting HCC progression. These findings indicate for the first time that serum SULF-2 could serve as a prognostic marker in patients with HBV-related HCC.

P04-01YI Hepatitis B core-related antigen quantification is an accurate predictor 12-months prior to hepatitis B "e" antigen-seroclearance in human immunodeficiency virus/hepatitis B coinfected patients treated with tenofovir

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Background and aims: Hepatitis B core-related antigen (qHBcrAg) can be a valuable tool in estimating viral activity and liver disease in patients chronically infected with HBV. This surrogate marker is well correlated with intrahepatic levels of HBV cccDNA, even during potent antiviral therapy. Nevertheless, no previous study to date has examined its relevance during HIV-HBV co-infection. This longitudinal study aimed to assess whether qHBcrAg is a predictor of hepatitis B “e” antigen (HBeAg)-seroclearance in HBeAg-positive HIV-HBV co-infected patients.

Method: 100 HIV-HBV-infected, HBeAg-positive patients initiating tenofovir (TDF)-containing antiretroviral regimen were prospectively followed. HBV-DNA and qHBcrAg were obtained at baseline and every 6-12 months. Hazard ratio (HR) assessing the association between qHBcrAg and HBeAg-seroclearance was calculated using proportional hazards regression and the sensitivity (Se) and specificity (Sp) of various qHBcrAg levels in predicting HBeAg-seroclearance was assessed using time-dependent ROC curves.

Results: At TDF-initiation, patients were a median 40.7 years old and 94% were male. Median CD4+ cell count was 408/mm^3 (IQR = 300-573). Median HBV-DNA was 6.26 $\log_{10}$IU/ml (IQR = 3.58-7.58) and median qHBcrAg 7.6 $\log_{10}$IU/ml (IQR = 7.0-8.1). Higher baseline qHBcrAg levels were independently associated with originating from low/moderate-HBV prevalent region (p = 0.04), shorter duration of antiretroviral therapy (p = 0.004), ALT >70 IU/ml (p = 0.009), AST >70 IU/ml (p = 0.02), and higher HBeAg quantification (p <0.001). After a median 5.3 years (IQR = 3.0-7.7) of TDF, HBeAg-seroclearance occurred in 31 patients (9 with seroconversion) within a median 30 months (IQR = 21-59). Baseline qHBcrAg <6.5 $\log_{10}$ IU/ml was a predictor of HBeAg-loss during follow-up (HR = 5.63; 95% CI = 2.67-11.88), after adjustment for lagged CD4+ cell count and antiretroviral treatment duration. qHBcrAg ≤6.5 $\log_{10}$ IU/ml and undetectable HBV-DNA were used as criteria to predict HBeAg-seroclearance at various time points during TDF-treatment, as shown in Table 1.

Conclusion: In co-infected patients, HBcrAg levels appears to have high sensitivity in predicting HBeAg-seroclearance 12-months prior to loss. Although specificity in predicting long-term HBeAg-seroclearance was low, it remained mostly higher than HBV-DNA detection.

Figure:

Table 1 . Quantifiable HBV markers in predicting HBeAg-seroclearance

Criteria	Classification Probabilities								
	M24		M36		M48		M72		M96
	Se/Sp		Se/Sp		Se/Sp		Se/Sp		Se/Sp
qHBcrAg <6.5 $\log_{10}$ IU/ml									
M12	0.77/0.66		0.78/0.67		0.72/0.69		0.68/0.75		0.59/0.87
M24			1/0.56		0.97/0.59		0.90/0.66		0.76/0.80
M36					1/0.39		1/0.46		0.86/0.42
HBV-DNA <60 IU/ml									
M12	0.79/0.62		0.68/0.62		0.72/0.65		0.56/0.33		0.49/0.28
M24			1/0.33		1/0.35		0.92/0.40		0.83/0.49
M36					1/0.16		1/0.20		0.92/0.23

P04-02YI Anti-hepatitis B core antibody titer and interleukin-10 quantification as predictors of hepatitis B "e" antigen-seroclearance in human immunodeficiency virus/hepatitis B coinfected patients treated with tenofovir

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Background and aims: Quantified anti-hepatitis B core antibody (qAnti-HBc) has been used to predict efficacy of antiviral therapy in mono-infected patients with hepatitis B “e” antigen (HBeAg)-positive chronic hepatitis B (CHB). Furthermore, interleukin-10 (IL-10) has close implications with the immune system and pathogenesis of CHB, making it a candidate marker for therapeutic response. We evaluated the predictive capacity of these markers on HBeAg-loss during treatment in HIV-HBV coinfection.

Method: 99 HIV-HBV-infected, HBeAg-positive patients initiating tenofovir (TDF)-containing antiretroviral treatment (ART) were prospectively followed. HBV-DNA, qAnti-HBc and IL-10 quantification were obtained at baseline and every 6-12 months. Hazard ratios (HR) assessing the association between qAnti-HBc or IL-10 and HBeAg-loss were calculated using proportional hazards regression and the sensitivity (Se) and specificity (Sp) for each marker in predicting HBeAg-loss was assessed using time-dependent ROC curves.

Results: At TDF-initiation, patients were a median 40.8 years old and 94% were male. Median CD4+ cell count was 409/mm^3 (IQR = 302-580). Median HBV-DNA was 6.42 $\log_{10}$IU/ml (IQR = 3.58-7.58), median qAnti-HBc 2.96 $\log_{10}$PEIU/ml (IQR = 1.10-3.97) and IL-10 -1.10 $\log_{10}$IU/ml (IQR = -1.25, -0.97). Higher baseline qAnti-HBc levels were independently associated with higher age (p <0.001), CDC stage A/B (p = 0.02), CD4+ cell count per 100 cells/mm^3 (p <0.001), and higher HBV DNA (p = 0.04). Only higher age was independently associated (p = 0.07) with higher baseline IL-10 levels. After a median 5.3 years (IQR = 3.0-7.8) of TDF, HBeAg-loss occurred in 30 patients within a median 32 months (IQR = 21-59). Baseline qAnti-HBc $\geq$4.0 $\log_{10}$PEIU/ml was a predictor of HBeAg-loss during follow-up (HR = 5.48; 95%CI = 2.23-13.49), after adjustment for HBV-infection duration, lagged CD4+ cell count, ART duration and HBV-DNA. However, baseline at IL-10 at any cut-off was not independently associated with HBeAg-loss after adjustment and hence was not considered further. Table 1 shows Se/Sp for qAnti-HBc and undetectable HBV-DNA used to predict HBeAg-seroclearance during TDF-treatment.

Conclusion: In coinfected patients, baseline qAnti-HBc levels appear to have high Sp in predicting HBeAg-seroclearance during follow-up. Baseline IL-10 is not an adequate marker for predicting HBeAg-loss.

Figure: Table 1. Quantifiable HBV markers in predicting HBeAg-loss

Criteria		Classification Probabilities									
		M24 *n* = 79		M36 *n* = 65		M48 *n* = 45		M72 *n* = 31		M96 *n* = 13	
	N	Se	Sp	Se	Sp	Se	Sp	Se	Sp	Se	Sp
qAnti-HBc at baseline (log_{10}PEIU/ml)											
≥3.0	99	0.81	0.49	0.69	0.49	0.74	0.51	0.75	0.56	0.68	0.67
≥4.0	99	0.47	0.77	0.38	0.78	0.41	0.79	0.35	0.81	0.31	0.87
≥4.1	99	0.47	0.81	0.38	0.81	0.41	0.83	0.35	0.85	0.30	0.94
HBV-DNA <60 IU/ml											
M12	99	0.79	0.62	0.67	0.62	0.72	0.65	0.57	0.67	0.49	0.72
M24	87			1	0.33	1	0.35	0.92	0.61	0.83	0.49
M36	79					1	0.16	1	0.20	0.92	0.23

P04-03 Serum HBV RNA and HBcrAg: new markers for predicting incomplete HBV DNA suppression after initiation of nucleoside therapy in HIV/HBV co-infected individuals

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Background and aims: Hepatitis B core-related antigen (HBcrAg) and HBV RNA are markers of transcriptional cccDNA activity and emerging new biomarkers in the management of chronic HBV. Data on their clinical utility in HBV/HIV co-infection are lacking. A proportion of HBV/HIV co-infected patients, adhering to dual active antiretroviral therapy (ART) with suppressed HIV, have persistent HBV replication. In this pilot study we aimed to measure HBcrAg and HBV RNA concentrations, in chronic HBV/HIV patients at initiation of tenofovir (TDF) based ART, focusing on prediction of incomplete HBV suppression as an end point.

Method: 30 chronic HBV/HIV patients, on TDF based ART, were identified retrospectively and classified into 2 groups: those with fully suppressed HIV (<50 copies/ml) and HBV (<20IU/ml) at week 48 of ART (group A, n = 15) and those with persistent HBV replication despite undetectable HIV at week 48 (group B, n = 15). Serum HBcrAg and HBV RNA were tested at baseline (initiation of TDF based ART). HBV RNA was measured by real-time PCR research assay Abbott Diagnostic (Butler E et al. Hepatology 2018) HBcrAg was measured by CLEIA assay (Fujirebio).

Results: There were no significant differences between groups A and B in sex, age, HBV genotypes, and baseline CD4 and baseline HBV DNA. There was a higher proportion of HBeAg+ patients in group B (87 v 47%, p <0.05) but more patients in group A (43 v 0%, p <0.05) achieved HBeAg loss by week 48. At baseline group B patients had higher HBsAg levels (4.53 v 3.99 $\log_{10}$ IU/ml, p <0.01), HBcrAg levels (7.00 v 5.10 $\log_{10}$ U/ml, p <0.001) and HBV RNA levels (6.23 v 3.82 $\log_{10}$ U/ml, p <0.05) compared to group A. A baseline HBcrAg cut-off value of 6.85 $\log_{10}$ U/ml had good predictive value for incomplete HBV suppression at 48 weeks (sensitivity 93%, specificity 80%, PPV 82% and NPV 92%) and the area under the ROC curve (AUROC) was good at 0.891 (0.764, 1.000; 95% CI). A baseline HBV RNA cut off value of 5.42 $\log_{10}$ U/ml had good predictive value for incomplete HBV suppression at 48 weeks (sensitivity 80%, specificity 80%. PPV 80% and NPV 80%) and the AUROC was good at 0.830 (0.639, 1.000; 95% CI).

Conclusion: In this small HBV/HIV cohort baseline HBcrAg and HBV RNA levels were higher in those who failed to suppress HBV DNA despite good adherence to HBV active ART. There may be a role for these biomarkers in risk stratification of co-infected patients warranting closer surveillance and management. A larger cohort is planned to enable evaluation of their combined predictive value with the aim to fully establish the clinical utility of HBcrAg and HBV RNA in this group.

P04-04YI Genetic variation in STAT4 is associated with treatment response to pegylated interferon in patients with chronic hepatitis B

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Background and aims: Signaling pathway in the *STAT4* gene play an essential role in interferon (IFN)-mediated antiviral effect. Recent data showed an association of rs7574865, a single nucleotide polymorphism (SNP) in *STAT4*, with treatment response to IFN-alpha in HBeAg-positive chronic hepatitis B (CHB). This study was aimed at investigating the role of this SNP in Thai individuals with CHB receiving pegylated interferon (PEG-IFN).
Method: A total 261 Thai patients (115 HBeAg-positive and 146 HBeAg-negative CHB) treated with 48-week PEG-IFN were recruited. Virological response (VR) at 48 weeks post treatment was defined as HBeAg seroconversion plus HBV DNA <2, 000 IU/ml for HBeAg-positive CHB and HBV DNA <2, 000 IU/ml for HBeAg-negative CHB. The SNP was analyzed by *Taq*Man probe PCR assay.
Results: The distribution of GG, GT and TT genotypes of rs7574865 in this cohort was 41.8%, 42.9% and 15.3%, respectively. There was no different in its distribution according to HBeAg status. Overall, patients with TT genotype, compared with non-TT genotype, achieved higher VR (64.3% vs. 30.5%; p <0.001) and HBsAg clearance (23.8% vs. 5.0%; p <0.001). There was the same trend in the HBeAg-positive group (VR, 52.4% vs. 30.9%; P = 0.077; HBsAg clearance, 23.8% vs. 6.4%; P = 0.028) and in the HBeAg-negative group (VR, 68.4% vs. 32.3%; P = 0.004; HBsAg clearance, 21.1% vs. 4.7%; P = 0.026). Moreover, patients with TT-genotype had significantly higher HBsAg decline than those with non-TT genotype at the end of follow-up.
Conclusion: These data suggest that SNP rs7574865 in the *STAT4* gene is a predictive factor of response to PEG-IFN therapy in Thai patients with CHB, regardless of baseline HBeAg status.

P04-05YI Evaluation of serum alanine aminotransferase level in treated chronic hepatitis B patients

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Background and aims: The American College of Gastroenterology (ACG) suggested a true healthy normal serum alanine aminotransferase (ALT) level ranges from 29 to 33 IU/L for males and 19 to 25 IU/L for females. However, upper limits of normal (ULN) for ALT level was defined as 50 IU/L for males and 40 IU/L for females by Chinese guideline. The aim of our study was to evaluate the ALT level in Chinese treated chronic hepatitis B (CHB) patients who had taken entecavir (ETV) for 78 weeks and gone liver biopsy.
Method: Patients with alcohol use, HCV, HAV, HEV, HIV, CMV and EBV infection were excluded. Finally, 409 Chinese treatment-naïve CHB patients received ETV therapy for 78 weeks. Among which, 268 patients had gone liver biopsy. Liver fibrosis and inflammatory were staged by Ishak score (F ≤2 absence of significant fibrosis, HAI ≤4 absence of significant inflammatory). Patients with negative HBV DNA (≤1000 IU/ml), normal body-mass index (BMI, <24 kg/m2), normal blood lipids (serum triglyceride concentration ≤1.7 mmol/L, serum total cholesterol concentrations ≤5.69 mmol/L, serum glucose levels ≤6.1 mmol/L) and normal liver biopsy (F ≤2, HAI ≤4) were included in ALT level evaluation.
Results: The distribution of ALT levels is shown in Table 1 and Table 2. With the increase of clinical and biochemical factors, the median values and maximum values of ALT decreased. The median and maximum ALT values for men with negative HBV DNA, normal BMI, normal blood lipid and normal liver biopsy were 19.5 ± 10.5 IU/L and 36 IU/L, whereas in women, the corresponding figures were 13 ± 6 IU/L and 20 IU/L. The maximum ALT values for men and women were lower than Chinese guideline and close to ACG guideline.
Conclusion: In this study, the ALT level was the lowest in Chinese treated CHB patients who had negative HBV DNA, normal BMI, normal blood lipids and normal liver biopsy, which was close to ACG guideline.

Figure:

Table 1. ALT levels under different clinical and biochemical factors for men (IU/L)

Variable	N	Median	Range	2.5%	95%	97.5%
Total	319	26 ± 15	10-134	12	54	68.3
Negative HBV DNA	310	26 ± 14.9	10-134	12	50.9	61.5
Negative HBV DNA, normal BMI and blood lipids	121	24.3 ± 15	11-80	12	48	59.9
Negative HBV DNA, normal liver biopsy	68	22 ± 12.9	11-55	11.7	43.1	54.2
Negative HBV DNA, normal BMI, blood lipids and liver biopsy	25	19.5 ± 10.5	12-36	12	35.1	-

Values are expressed as the median ± interquartile range

Table 2. ALT levels under different clinical and biochemical factors for women (IU/L)

Variable	N	Median	Range	2.5%	95%	97.5%
Total	90	19 ± 11	5-54	9	42.5	50.7
Negative HBV DNA	84	19 ± 11	5-54	9	45.5	51
Negative HBV DNA, normal BMI and blood lipids	46	18.4 ± 11.2	9-54	9	50.5	53.5
Negative HBV DNA, normal liver biopsy	17	13 ± 6.9	9-29	9	-	-
Negative HBV DNA, normal BMI, blood lipids and liver biopsy	11	13 ± 6	9-20	9	-	-

Values are expressed as the median ± interquartile range

P06-01YI Hepatitis B virus infection as a trigger of hepatocyte identity loss

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Background and aims: Human hepatocytes are the natural host of Hepatitis B virus (HBV) infection. HBV is able to persist within hepatocytes as a covalently closed circular DNA (cccDNA), inducing hepatitis, cirrhosis and hepatocellular carcinoma (HCC). HCC is the result of alterations of cellular processes due to chronic infection, the oncogenic properties of viral proteins such as HBx, the host immune response, and the effect of a chronic inflammatory microenvironment. This combination of factors leads to the loss of hepatocyte identity, due to arrested differentiation or de-differentiation.
At the molecular level, hepatocyte identity is dependent on the action of transcription factors and the epigenetic machinery; Hepatocyte Nuclear Factor-4α (HNF4A) is a master orchestrator of such identity. The *HNF4A* gene codes for several isoforms that originate from two different promoters. Previously, we have shown that there is a drastic induction of *HNF4A* promoter 1 (P1) isoforms after one week of progenitor differentiation towards hepatocytes. This is followed by progressive downregulation of P2-dependent isoforms, in a fashion that we propose acts as an epigenetic switch. The present study searches for an explanation for the loss of hepatocyte identity observed after HBV infection. Our specific aims are to characterize the effect of *in vitro* HBV infection on hepatocyte (de) differentiation and to understand the specific role DNA methylation in this process.
Method: A differentiation protocol of 4 weeks using HepaRG cells was employed to obtain mature hepatocyte like cells. Cells were naturally infected with HBV (500MOI); Total HBV, cccDNA, HBs and HBe were quantified to validate infection efficacy. The methylation status of HNF4A promoters were elucidated by qMSP and the functional effects of HBV infection were determined by qPCR and Western Blot.
Results: A sustained infection was achieved after 8 days. Differential expression of players in the HNF4A epigenetic switch were observed by qPCR in cells exposed to HBV; including, a decrease in expression of HNF4A P1 isoforms and FOXA2 as well as changes in the methylation of HNF4A promoters in cells exposed to HBV. More work is needed to elucidate the mechanisms by which these changes occurred, however, our work indicates that the HNF4A epigenetic circuit is implicated in the differentiation status of hepatocytes exposed to HBV.
Conclusion: Deregulation of the HNF4A identity circuit by HBV may be established as a key mechanistic event in the pathway towards HBV-related chronic liver disease. Moreover, early epigenetic events triggered by HBV may represent useful biomarkers of cancer risk.

P06-02YI Serum M2BPGi as a diagnostic marker of liver fibrosis in patients with chronic hepatitis B

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Background and aims: Serum M2BPGi represents a novel biomarker of liver fibrosis, particularly in patients with chronic hepatitis C. The aim of this study was to evaluate the diagnostic role of this serum marker in patients with chronic hepatitis B (CHB).
Method: Serum M2BPGi levels were measured by an automated lectin-antibody sandwich immunoassay in stored serum samples collected at initial diagnosis of consecutive 410 patients with CHB (without hepatocellular carcinoma). Liver stiffness measurement (LSM) was assessed by transient elastography.
Results: Among the initial enrollment, 14 patients with invalid LSM and 91 patients with an increased serum ALT level ≥2×upper limit of normal (ULN) were excluded. Thus, there were 305 patients for further analysis. Based on LSM, patients with fibrosis stage F0-F1, F2, F3, and F4 were 120 (39.3%), 93 (30.5%), 45 (14.8%) and 47 (15.5%), respectively. The median values of M2BPGi in the corresponding fibrosis stages were 0.9, 1.1, 1.4 and 1.8 cutoff index (COI), respectively. The ROC curves analyses were performed for significant fibrosis (≥F2), advanced fibrosis (≥F3) and cirrhosis (F4). Our data showed that the area under ROC curve for diagnosis of the corresponding fibrosis stages were 0.76 [95% confidence interval (CI); 0.72-0.83, p <0.001], 0.80 (95% CI; 0.75-0.85, p <0.001) and 0.86 (95% CI; 0.82-0.91, p <0.001). The optimal cut-off values were 1.4 COI for ≥F2, 1.6 COI for ≥F3 and 1.9 COI for F4. The sensitivity of M2BPGi for the corresponding fibrosis stages was 63.2%, 70.7% and 83.0%, the specificity was 66.7%, 61.5% and 76.7%, while the accuracy was 66.7%. 70.4% and 82.0%.
Conclusion: Serum M2BPGi levels progressively increased with the severity of liver fibrosis in patients with CHB. Thus, serum M2BPGi can be severed as a useful non-invasive marker for liver fibrosis, particularly in patients with cirrhosis.

P06-03YI Association between Hepatitis B Virus preS2 deletion and increased risk of Hepatocellular Carcinoma: a case-control study in West Africa

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Background and aims: Hepatitis B virus (HBV)-related HCC (Hepatocellular Carcinoma) is endemic in West Africa (WA). Previous results from the PROLIFICA (Prevention of Liver Fibrosis and Cancer in Africa) (EU FP7) project have shown that individuals in WA have low viral loads (2, 000 UI/ml) (Ghosh et al., 2016), an early onset of cirrhosis and a high occurrence of liver tumours (Lemoine et al., 2016). In WA exposure to Aflatoxin B1 (AFB1), a carcinogenic mycotoxin contaminating food also associated with increased HCC risk, is also commonplace. As AFB1 exposure has been shown to reduce HBV replication *in vitro* (Lereau et al., 2012), our study aimed at investigating potential links between AFB1 exposure, HBV parameters *in vivo* and HCC risk.
Method: HBsAg-positive individuals with or without cirrhosis or HCC from the population-based screening program, and HBV-related cirrhotic cases and HCC from the referral hospital were recruited in West Africa. Comprehensive clinical and virological assessments were performed, together with *in vitro* characterization of mutant HBV strains. HBV variants were detected using PCR, Sanger sequencing, next-generation sequencing and cloning. AFB1 exposure was assessed using the R249S *TP53* mutation in circulating free DNA as a surrogate biomarker and detected by next-generation-sequencing. Multiple sequence alignments were done using MAFFT and phylogenetic trees inferred using FigTree (Maximum Likelihood, GTR+G4+I). HepaRG and HepG2-NTCP cell lines and Primary Human Hepatocytes were infected with HBV and/or exposed to AFB1 *in vitro* and viral markers assessed.
Results: This study population was mainly infected with HBV genotypes A and E (85 and 15%, respectively) with deletions in the preS2 domain frequently being detected. This HBV mutation profile differs from that seen elsewhere as it is centred on bps 412-429 of the surface gene and varies in length from 12 to 18 bps. Based on a phylogenetic analysis (n = 302) the deletions appear to arise during the natural course of the infection in individuals, are potentially transmissible and their frequency significantly increases with disease progression (23% of chronic HBV carriers, 42% of cirrhotic patients and 57% of HCC patients, p = 0.01). 18% of the 155 individuals analysed carried this AFB1 associated *TP53* mutation confirming the exposure of the study population to this carcinogen. *In vitro* data confirm that AFB1 exposure reduces HBV infection parameters (DNA, RNA and HBsAg and HBeAg levels) and these studies are now being extended to the HBV genotypes found in WA.
Conclusion: HCC in this study population is associated with (1) the carriage of a preS2 HBV deletion mutations that are localised in one specific region of the preS2 gene and (2) AFB1 exposure. However, the crosstalk between these factors and the replication of different HBV genotypes on HCC risk remain to be fully deciphered.

P06-04YI The effect of diabetes mellitus on treatment course of chronic hepatitis B and their clinical profile

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Background and aims: Natural history of hepatitis B in the middle east is lacking. We aim to assess the natural history and clinical characteristic profile of patients with positive HBsAg and the effect of diabetes mellitus in our cohort, and to evaluate diabetes and other comorbidities burden in these patients, and to further understand the outcomes resulting from different treatment regimens.
Method: Cross-sectional chart-review of all hepatitis B database in King Fahad Specialist Hospital-Qassim-Saudi Arabia. The Database includes all patients who underwent treatment for hepatitis B in the hospital. Data analysis was done using the SPSS program version 20.
Results: A total of 133 patients were included in this study. The mean follow-up time was 4 years and the mean age was 45.2 ± 14.4 years. Out of total 62.4% (83) patients were males and 37.6% (50) were females. 19 patients were cirrhotic, 10 Child-Pugh A, 4 Child-Pugh B and 4 had Child-Pugh C. 15% (20) had diabetes mellitus and 12% (16) had hypertension. 14.3% (19) HBeAg +, and during the follow-up period, two of them had sero-conversion to HBeAg negative state. One patient after 7 years of treatment with multiple anti-viral drugs and the other case after two years of treatment with Tenofovir. Entecavir was chosen as a first-line drug in most of the patients 70.7% (94), followed by Tenofovir 13.5% (18). 8.3% (11) had not responded at some time during their follow-up in the hepatology clinic. Five of them had a positive HBeAg status, one had cirrhosis, and one was diabetic. The reasons for non-response were non-compliance and resistance. Resistance was found to Entecavir, Adefovir, lamivudine, PEG-interferon as a monotherapy and combination of them. Tenofovir was used as salvage therapy.
Conclusion: Most of Chronic hepatitis B are HBeAg negative. Response to oral nucleoside/nucleotide analogues therapy in complaint patients are optimum in both cirrhotic and non-cirrhotic patients. 15% of our cohort had Diabetes Miletus. The course of chronic hepatitis B and response to treatment was similar in both diabetic and non-diabetics.

P06-05 Mean Week 24 HBsAg declines predict subsequent rate of HBsAg seroclearance and could be a valuable end point for early development hepatitis B virus trials

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Background and aims: Early identification of potentially efficacious novel hepatitis B virus (HBV) treatment modalities in clinical development requires on-treatment response markers. We hypothesized that on-treatment mean HBsAg decline could serve as a marker for subsequent off-treatment HBsAg seroclearance at the treatment arm level.

Method: We used two complementary approaches to investigate the relationship between mean HBsAg decline at Week 12 or 24, and subsequent HBsAg seroclearance at either Week 48 of treatment or 24 weeks after end of treatment: (1) A model-based meta-analysis based on a systematic review of published studies (40 treatment arms/cohorts) of pegylated Interferon alpha (PEG-IFN) and nucleos (t)ide analogue (NA) treatment; and (2) A mixture model consisting of a within-study comparison of HBsAg decline between patients with and without HBsAg clearance. This second approach used individual patient data from: the HBV 9901 study (PEG-IFN versus PEG-IFN with lamivudine for HBeAg-positive patients with chronic hepatitis B (CHB); the PARC study (PEG-IFN alone or with ribavirin for HBeAg-negative CHB); and published data from Study 0149 (PEG-IFN with or without tenofovir for HBeAg-positive and -negative CHB) and LIRA-B (PEG-IFN for HBeAg-positive CHB).

Results: The model-based meta-analysis and mixture modelling showed consistent results. Across trials, a more pronounced mean HBsAg decline at Week 24 was associated with higher rates of subsequent HBsAg seroclearance. A mean HBsAg decline at Week 24 of 0.5 $\log_{10}$ can be considered a minimum threshold for the occurrence of subsequent HBsAg clearance. Also, an additional mean 1 $\log_{10}$ decline at Week 24 compared to a comparator treatment arm would be expected to translate into a 20 to 30% increase in subsequent HBsAg seroclearance. For Week 12 data, similar observations were made but the relationship was less strong.

Conclusion: Mean Week 24 HBsAg decline can predict subsequent HBsAg loss and an additional mean 1 $\log_{10}$ decline compared to a comparator arm is projected to translate to a 20 to 30% increase in subsequent HBsAg seroclearance on a treatment arm level. Mean Week 24 HBsAg decline could therefore be used as a valuable end point for early decision making in HBV clinical trials. While it is likely that this relationship also holds for new immunomodulators and direct acting antivirals, it is unlikely that this is true for drugs that directly interfere with HBsAg production, such as siRNAs.

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