HBsAg clearance during transaminase flares predicts control of HBV infection during NAP-based therapy of HBV or HBV / HDV infection

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INTRODUCTION

Transaminase flares during treatment of chronic HBV infection indicate the death / clearance of hepatocytes[1]. They occur with varying degrees and frequency with all approved therapies for chronic HBV infection[2-7]. Except during strong viral flares, transaminase flares are rarely associated with signs of hepatotoxicity and usually associated with improved virologic status or response to treatment. Transaminase flares are most common during treatment of HBV or HBV / HDV infections with pegylated interferon and may be associated with HBsAg loss and functional cure.

NAPs (i.e. REP 2139) selectively block the assembly and secretion of HBV subviral particles (SVP) (see poster adjacent and[8]), blocking replenishment of HBsAg and allowing its clearance by the host immune system. This effect appears to be important for the improved activity of various immunotherapies[9,10]. In addition to this mechanism, a separate upstream direct acting activity of REP 2139 against HDAg[11] may play a role in the complete clearance of HDV RNA in human participants.

In the REP 301 (HBV / HDV) and REP 401 (HBV) trials, a total of 52 participants with chronic HBeAg negative infection were exposed to REP 2139 and pegylated interferon (pegIFN), yielding high rates of HBsAg loss, HBsAg seroconversion and persistent control of both HBV and HDV infection after removal of all therapy. These effects were accompanied by transaminase elevations > upper limit of normal (ULN) in 96% of participants. HBV outcomes after removal of all therapy were rebound (29%), establishment of inactive chronic HBV / functional control (HBV DNA ≤ 2000 IU/mL, normal ALT) (38%) and establishment of functional cure (HBsAg < LLOQ, HBV DNA target not detected, normal ALT) (33%).

The goal of this analysis was to characterize the nature of the transaminase flares observed in these studies and their relationship to HBV therapeutic outcome.

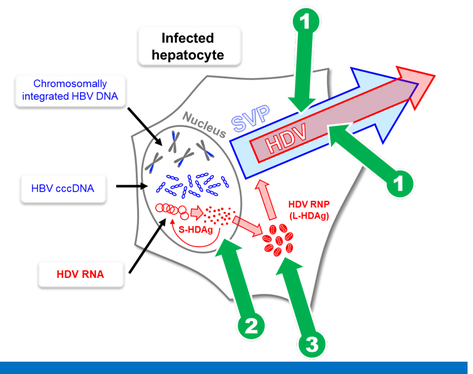


Figure 1. Antiviral effects of REP 2139:

(1) Inhibition of HBV SVP and HDV assembly / secretion.

(2) Potential inhibition of HDV RNA synthesis via interaction with S-HDAg.

(3) Potential inhibition of HDV RNP formation via interaction with L-HDAg.

METHODS

A retrospective analysis of all 52 participants from the REP 301 and REP 401 studies included comparison of maxima / mimima and area under the curve for ALT, AST, GGT, bilirubin, albumin and INR during exposure to 48 weeks of pegIFN with baseline virologic characteristics, HBsAg response during flare activity and its relationship to therapeutic outcome (48 weeks – 2 years follow-up) were also analyzed.

RESULTS

Table 1. Baseline characteristics according to HBV therapeutic outcome.

Group		Rebound (n=15)	Functional control (n=20)	Functional cure (n=17)	p-value
Age ($\bar{x}$ / median)		39.5 / 44	37.2 / 37	37.7 / 39	0.750[b]
Sex	Male	12	16	14	0.979[c]
	Female	3	4	3	
HBV genotype	A	1	1	0	0.922[c]
	D	10	14	14	
	ND[a]	4	5	3	
Baseline HBsAg (IU/mL, $\bar{x}$ ± SD)		$1.67x10^4$ ± $8.23x10^3$	$1.12x10^4$ ± $7.03x10^3$	$1.13x10^4$ ± $1.38x10^3$	0.274[b]
Baseline HBV DNA (IU/mL, $\bar{x}$ ± SD)		$9.66x10^6$ ± $2.28x10^7$	$3.73x10^6$ ± $1.57x10^7$	$3.46x10^7$ ± $1.40x10^8$	0.494[b]
Baseline transaminases (U/L, $\bar{x}$ ± SD)	ALT	76.1 ± 69.2	96.4 ± 72.4	88.4 ± 66.0	0.695[b]
	AST	46.5 ± 30.8	57.0 ± 38.0	51.9 ± 25.6	0.635[b]
	GGT	31.4 ± 24.6	33.1 ± 16.4	39.6 ± 25.6	0.531[b]
Baseline LMS (kPa)	≤ 7	6	10	7	0.942[c]
	7 - 9	5	5	5	
	9 - 11	1	3	0	
	11 - 18	2	2	3	
	>18	0	0	2	
Transaminase flare during therapy (> 3x ULN)	ALT	11	16	14	0.733[c]
	AST	6	9	13	
	GGT	4	10	11	
	Any	11	17	17	0.248[c]
HBsAg seroconversion (≥ 10 mIU/mL)		4	14	16	< 0.001[c] (0.558[d])

[a] Not determined; baseline HBV DNA too low for genotype determination due to HDV co-infection.
[b] Significance of variance of means between groups was determined by single factor ANOVA.
[c] Significance of variance of distribution of parameters between groups was determined by X^2 test.
[d] X^2 test for difference between functional control and functional cure groups only

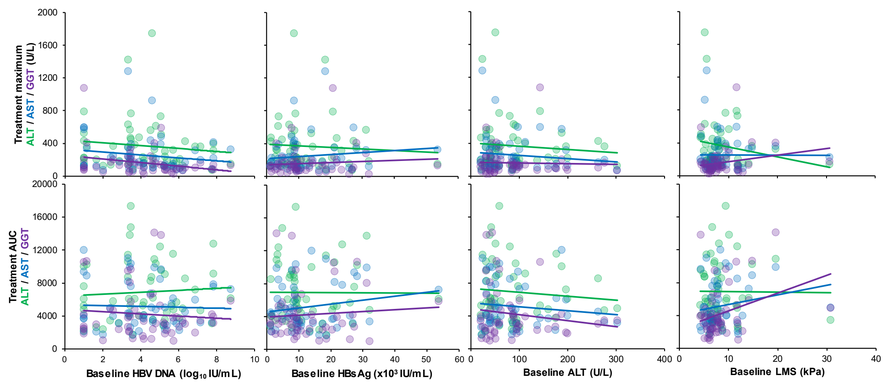
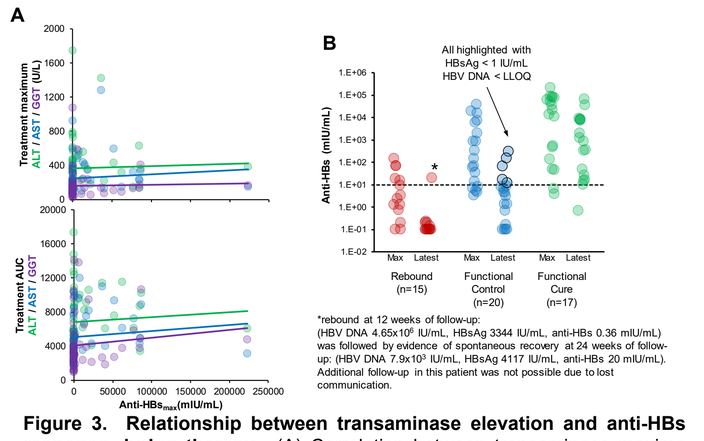


Figure 2. Relationship between transaminase elevation and baseline virologic status. Regression analysis of transaminase maxima (top) or AUC during therapy (bottom) are presented for ALT (green), AST (blue) and GGT (purple) versus baseline HBV DNA (left), HBsAg (center left), ALT (center right) and baseline liver median stiffness or LMS (right). No statistically significant correlations were observed .



*rebound at 12 weeks of follow-up: (HBV DNA $4.65x10^6$ IU/mL, HBsAg 3344 IU/mL, anti-HBs 0.36 mIU/mL) was followed by evidence of spontaneous recovery at 24 weeks of follow-up: (HBV DNA $7.9x10^3$ IU/mL, HBsAg 4117 IU/mL, anti-HBs 20 mIU/mL). Additional follow-up in this patient was not possible due to lost communication.

Figure 3. Relationship between transaminase elevation and anti-HBs response during therapy. (A) Correlation between transaminase maxima (top) and AUC (bottom) for ALT (green), AST (blue) and GGT (purple) with anti-HBs titer during therapy. (B) Anti-HBs maxima achieved during therapy (Max) and at the latest published follow-up visit (Latest) in participants in the different outcome groups. Dashed line indicates threshold for seroconversion (10 mIU / mL). Seroconversion is only maintained with functional cure or functional control where HBsAg < 1 IU/mL and HBV DNA < LLOQ.

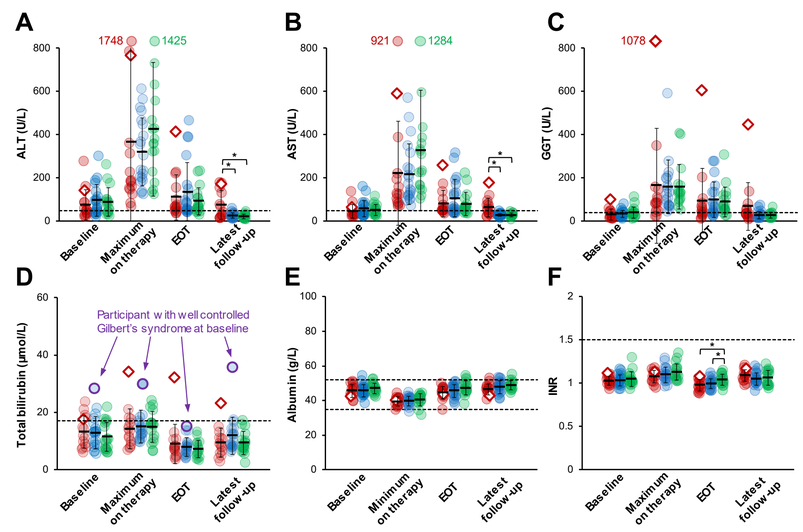


Figure 4. Transaminase elevations occur regardless of outcome, and do not effect liver function. Alterations in ALT (A), AST (B), GGT (C), total bilirubin (D), albumin (E) and INR (F) at baseline, maximum / minimum during therapy, end of therapy (EOT) and at the latest available follow-up are presented. Average and standard deviation are presented for rebound (red), functional control (blue) and functional cure (green). Statistically significant differences between specific outcomes (determined by T-test) are indicated by asterisks. Dashed horizontal lines indicate normal ranges or ULN. Note GGT was not assessed beyond 24 weeks during follow-up in REP 401 participants. The single DILI observation (in the REP 301 study) is indicated by a hollow diamond.

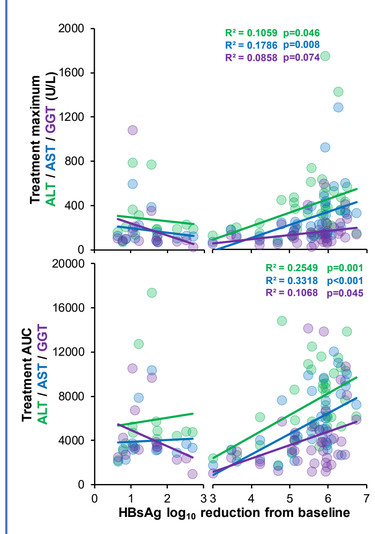
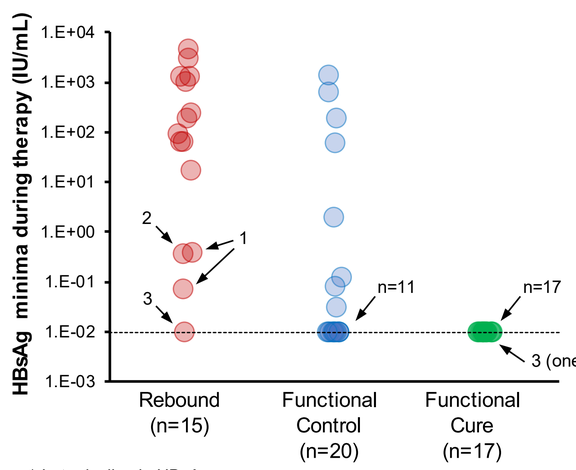


Figure 5. Relationship between transaminase flare activity and HBsAg reduction from baseline. Regression analysis of transaminase maxima (top) and AUC during therapy (bottom) are presented for ALT (green), AST (blue) and GGT (purple) versus HBsAg reduction from baseline. Correlation coefficients and statistical significance (determined by regression ANOVA) for each transaminase are indicated in the top right of each graph.



1. Late decline in HBsAg.
2. Early withdrawal from therapy due to pegIFN related depression.
3. Early withdrawal from therapy due to personal reasons not related to tolerability.

Figure 6. Relationship between HBsAg minima achieved during therapy and therapeutic outcome. All participants experiencing functional cure achieved HBsAg target not detected (0.00 IU/mL). All rebound participants experiencing HBsAg < 10 IU/mL either had very late decline in HBsAg of halted therapy early (see notes above). Dashed line indicates target not detected (0.00 IU/mL).

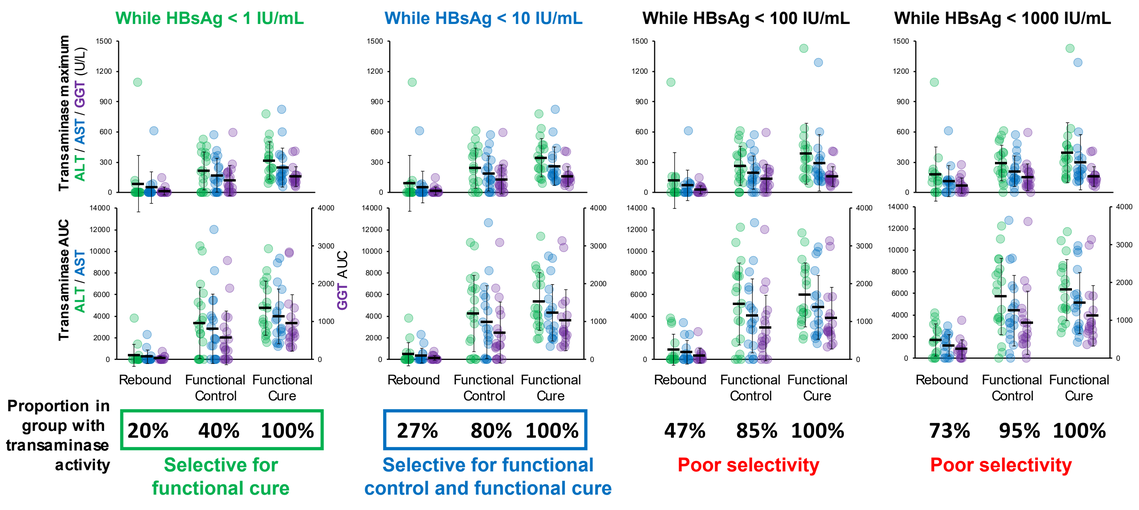


Figure 6. Extent of HBsAg clearance during transaminase activity predicts functional control and functional cure. Transaminase maxima (top) and AUC (bottom) during various thresholds of HBsAg clearance are indicated in different outcome groups for ALT (green), AST (blue) and GGT (purple). Average and standard deviation are indicated for each group. In all cases flare activity was significantly elevated in participants achieving functional control and functional cure. Only transaminase elevations present during the most efficient clearance of HBsAg appeared to have predictive value for functional control or cure (HBsAg < 10 IU/mL) or specifically for functional cure (HBsAg < 1 IU/mL)

CONCLUSIONS

- Transaminase flares are highly prevalent during NAP / pegIFN combination therapy but are well tolerated and except in one case, not associated with any signs of liver dysfunction.
- Flares are independent of baseline characteristics or therapeutic outcome but are positively correlated with HBsAg reduction > 3 log_{10} from baseline.
- HBsAg seroconversion is independent from flare activity and similar between functional control and cure. Persistence of seroconversion during functional cure suggests control of HBsAg synthesis.
- Improved HBsAg clearance during flares better predicts control of infection off therapy, with HBsAg < 1 IU/mL having the best association with functional cure.
- At all HBsAg thresholds examined, flares were significantly stronger in the functional control / cure groups versus rebound group, indicating stronger flares may have a positive prognostic value.
- **HBsAg-specific immunity may play an important role in restoring functional cure, an activity which may be efficiently potentiated only when HBsAg is < 1 IU/mL.**

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