INVITED REVIEW

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The role of HBsAg levels in the current management of chronic HBV infection

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Abstract

Chronic hepatitis B virus (HBV) infection can result in liver cirrhosis, hepatic decompensation, and hepatocellular carcinoma (HCC). However, the natural course of the disease is highly dynamic and not every patient requires therapy. The challenges for optimal management are who to treat, which therapeutic regimen to use, and when to begin or stop treatment. Constant monitoring is mandatory to predict the natural course and guide treatment decisions. Surrogate markers for baseline and on treatment decisions are needed. Besides HBV DNA, hepatitis B surface antigen levels also proved to be useful to help judge the natural course and guide treatment. High levels of HBsAg are suggestive of low fibrosis and immune tolerance in hepatitis B e antigen (HBeAg) positive patients; whereas low levels of HBsAg indicate a lower risk for HCC and inactive carrier state in HBeAg negative patients. Data also support the possible use of HBsAg levels as an on-treatment response marker. So far, the best evidence exists for treatment with interferon (IFN)-α where lack of HBsAg decline after 12 weeks is associated with non-response. Thus, stopping rules after 12 weeks therapy could be established for HBeAg positive as well as for HBeAg negative patients. However, the positive predictive value for achieving sustained response is still vague. The value of HBsAg monitoring is less clear during treatment with nucleos(t)ide analogues (NA) but it can be a useful marker for new concepts such as stopping NA or add-on IFN strategies. Currently, several studies are underway to validate HBsAg in these settings.

Keywords Chronic hepatitis B, hepatitis B surface antigen, interferon α, nucleos(t)ide analogues

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Introduction

Hepatitis B virus (HBV) is one of the world's most prevalent virus infections with about 350 million people chronically infected and about 600,000 hepatitis B-related deaths annually [1,2]. In this regard, as one of the world's most dreaded virus infections, HBV shares many similarities with the human immunodeficiency virus (HIV) and the hepatitis C virus (HCV). After closer examination however, there are substantial differences between these viruses. For HCV it is possible with present and upcoming new treatments to clear the virus completely and achieve a sustained virological response (SVR) [3]. For HIV, in contrast, a lifelong virus suppression therapy is mandatory, as there is no current option to clear the virus from host cells. HBV is different to the aforementioned viral infections as, due to cccDNA, a complete clearance from host cells is not feasible, but, in contrast to HIV infection, induction of immune responses and control of the virus is possible in many patients.

Chronic HBV-infection is distinguished in different phases. The phases are differentiated by HBV DNA, hepatitis B e antigen (HBeAg) and its antibody (anti-HBe), hepatitis B surface antigen (HBsAg) and its antibody (anti-HBs), respectively. The phases are the immune-tolerant phase, the immune-active phases, which require treatment in most cases, and the HBeAg negative-inactive carrier state, in which treatment is not necessarily needed [4]. Some HBV variants have mutations in the precore (PC) and basal core promoter (BCP) regions of their genome [5]. These variants escape from the anti-HBe antibody response. The consequence is HBeAg negative hepatitis with an increased risk for disease progression. However, it is sometimes challenging to discriminate active from inactive HBeAg negative chronic HBV infection.

As HBV cannot be eliminated, the ultimate aim for the

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treatment of HBV-infection is the loss of HBsAg and anti-HBs seroconversion, which indicates control of the disease by the immune system without need for further medication. Unfortunately this goal is rarely reached. Hence, ongoing surveillance of chronic HBV infection is mandatory, as are constant decisions about treatment choices.

As not all phases of chronic HBV infection require treatment, guidance is needed to help determine when to initiate treatment, terminate treatment and predict a treatment response.

In the past, HBsAg has been primarily used to diagnose HBV infection. In recent years two commercially available assays for HBsAg quantification, the Architect HBsAg assay (Abbott Diagnostics, Abbott Park, IL, USA) [6] and the Elecsys HBsAg II quant assay (Roche Diagnostics, Indianapolis, IN, USA) [7] have been developed. Results from both assays are strongly correlated (r>0.96), so obtained results are not dependent on the used platform [8,9]. Furthermore, recent data shows that HBsAg reflects the transcriptional activity of the cccDNA and that HBsAg is useful as a surrogate marker to predict the natural course of HBV-infection, HBV-coinfection and for treatment guidance [10].

HBsAg and natural course

The natural course of perinatally acquired chronic HBeAg positive chronic HBV-infection can be distinguished into different phases, which will be described in further detail with an emphasis on HBsAg, which may be useful in the differentiation (Table 1).

Perinatally acquired HBV-infection starts with the immune tolerant phase, which lasts for about 20-30 years. This phase is characterized by high HBV DNA levels and normal transaminases. Recent studies showed that HBsAg level tend to be about 4.9 log IU/mL for Europe and Asia [11,12]. HBsAg values of >100,000 IU/mL are suggestive for immune tolerance [13].

Patients in the immune tolerance phase usually show no or minimal histological lesions. A recent study showed that in HBeAg positive patients with low alanine aminotransferase (ALT), a HBsAg level >25,000 IU/mL had a >90% positive predictive value (PPV) for liver fibrosis <F1 (ISHAK-Score) [14]. These findings could reduce the need for liver biopsy and guide possible treatment decisions. In line with these results, Martinoux-Peignoux *et al* demonstrated that lower HBsAg levels in HBeAg positive patients but not in HBeAg negative patients were associated with moderate to severe fibrosis [15]. These findings indicate that HBsAg levels in HBeAg-positive patients have to be interpreted differently from HBeAg negative patients and may reflect the already partial control of the immune system in HBeAg negative patients.

The immune tolerant phase is followed by the immune clearance phase, in which the immune system tries to eliminate the virus. This phase is characterized by variable HBV DNA levels, which in general tends to decrease. On liver biopsy, there is marked active hepatitis. ALT levels are fluctuating, but tend to be above the upper normal limit. So far, there is no evidence that HBsAg levels help to predict anti-HBe seroconversion [13]. Nevertheless, HBsAg levels drop and reflect the ongoing immune response. In most patients, the immune clearance phase results in anti-HBe seroconversion, but some patients do have an unsuccessful immune clearance. These patients are at increased risk for development of cirrhosis and/or hepatocellular carcinoma (HCC).

The best usual scenario for HBsAg positive patients is the development of HBeAg negative inactive carrier state, where anti-HBe antibodies are present, HBV DNA is low, and there are no marked signs of hepatitis in the liver biopsy. Because of the fluctuating ALT and HBV DNA levels, the differentiation between ongoing HBeAg negative chronic hepatitis and the inactive carrier state may be difficult and requires close monitoring [4,16,17].

To differentiate the latter two phases and estimate the prognosis of the natural course of HBV infection, different markers have been studied. The REVEAL Study showed that the development of HCC depends on the viral load, measured by serum HBV DNA. HBV DNA of >2,000 IU/mL is associated with a cumulative incidence of HCC of 3.57% after 13 years. The risk becomes more prominent with rising viral loads and surpasses 12% for HBV DNA levels exceeding 20,000 IU/mL [18].

Hence, the current recommendations for the American and European guidelines use the HBV DNA cut-off of 2,000 IU/mL to define the inactive carrier state, which is correlated with good survival prognosis and low incidence of cirrhosis and HCC [4].

Further help to discriminate between inactive carriers and chronic HBeAg negative hepatitis B can be provided by assessing the HBsAg levels. Several studies showed that inactive carriers had lower HBsAg levels [11,19,20], with HBsAg levels <1,000 U/mL having a 87.9% PPV and 96.7% NPV for patients with HBV DNA <2,000 IU/mL [21].

In line with the HBsAg cut-off of <1,000 IU/mL, the REVEAL study demonstrated that HBsAg level of <1,000 IU/mL and even more <100 IU/mL is linked to a lower risk for HCC development in the predominant HBeAg negative study population [22].

The ultimate goal in HBV infection is HBsAg loss. Patients who underwent anti-HBs and anti-HBe seroconversion had the absolute lowest risk with a cumulative risk for HCC <1% over 9 years, which shows the huge importance of HBsAg seroconversion and immune control of the virus [18]. Fattovich *et al* showed that patients with compensated cirrhosis with HBsAg loss had a survival of nearly 100% after 14 years [23]. As a major cofactor to low HBV DNA for anti-HBs seroconversion, HBsAg levels of <100 IU/mL at baseline corresponded to an increased chance of HBsAg clearance [24], but further evaluation is needed.

In this regard, attention should be focused on the specific HBV genotype, as highest levels of HBsAg are found in genotype A infected patients [25], with genotype A and D having overall higher values than genotype B and C [11]. Neglecting these facts could lead to over- and under-estimation of future HBsAg cut-offs in distinct patient populations.

Treatment

The goals for treatment for chronic hepatitis B are, as defined by the EASL in 2012 [4], HBsAg loss and sustained HBV DNA suppression. These parameters can both be measured and are, as outlined in the paragraph on the natural course of the infection, strongly correlated to good survival and prevention of complications associated with HBV-infection, i.e. cirrhosis and HCC.

The critical step required to achieve HBsAg loss is sustained immune control of the virus. This step is defined by durable anti-HBe seroconversion with HBV DNA <2,000 IU/mL for HBeAg positive HBV infection or a sustained HBV DNA <2,000 IU/mL and normal ALT for HBeAg negative HBV infection in the long term [26,27].

To date, there are 2 classes of agents for treatment of chronic hepatitis B: interferon (IFN)-α and nucleoside or nucleotide analogues (NA).

IFN-α treatment

The aim of treatment with IFN is to achieve finite immunologic control and HBsAg clearance after therapy. Hence, IFN therapy is a time-restricted therapy, usually given for 48 weeks [4]. Its purpose is sustained off-treatment response with low virus load and normal ALT without further required treatment. In comparison with the NA-treatment, pegylated IFN (PEG-IFN) produces rates of immunological response of about 30% anti-HBe seroconversion [28] after 6 months of treatment, whereas the rate of anti-HBe seroconversion is slightly lower (approximately 20% after 12 months of NA-treatment) [4]. In addition, NA induced anti-HBe seroconversion seems not to be as durable compared to IFN, showing higher relapse rates [29]. However, the tolerability of IFN is lower compared to NA. Thus, baseline as well as on-treatment predictive markers are urgently needed to decide which patients benefit from a finite IFN treatment.

Several studies have been published to identify markers to guide the treatment with baseline predictors for response. As mentioned before, anti-HBe seroconversion is defined as one of the markers of immune control and correlated with a significantly better survival and markedly reduced incidence of cirrhosis [30]. In the NEPTUNE Study, 61% of patients with an ALT >5 upper limit of normal (ULN) at baseline achieved anti-HBe seroconversion 6 months after PEG-IFN treatment for 48 weeks, whereas seroconversion was observed in only 19% of patients with ALT <2 ULN [31].

Similar data can be shown for the HBeAg negative chronic hepatitis B, in which patients with increased ALT had an increased chance of a virological response [32]. The HBV genotype and the presence of PC and BCP mutations may be also relevant for baseline prediction of treatment response to IFN. As Sonneveld *et al* reported, patients with genotype A and wild-type virus at baseline have the greatest chance for successful anti-HBe seroconversion, whereas pre-existing PC and BCP mutations in HBeAg positive patients were correlated with reduced response rates to IFN [33].

Some data suggest that lower HBsAg levels at baseline are associated with a better treatment response [34]. However, HBsAg levels at baseline depend on the phase of HBV infection and the genotype [11]. For example, HBeAg positive patients, who were non-responders to IFN therapy, showed notable differences in their HBsAg kinetics across the different genotypes A to D [35]. This supports the above mentioned notion that HBsAg levels need to be validated for a distinct genotype.

Current data supports the further use of HBsAg as an on-treatment surrogate marker for sustained treatment response in HBeAg positive chronic hepatitis B (Table 2). Early treatment response with HBsAg levels <300 IU/mL after 6 months of treatment is correlated with a sustained response defined as anti-HBe seroconversion and HBV DNA <2,000 IU/mL until 12 month post-treatment (62% vs 11%) [36]. Likewise, in the phase III study of PEG-IFN-2α, HBsAg levels of <1,500 IU/mL at week 12 and week 24 had PPV of 57% and 54% for anti-HBe seroconversion. The rate of HBsAg clearance was 17.6% in patients who had HBsAg level <1,500 IU/mL at week 12 [37].

In contrast, patients who did not experience a significant HBsAg decline at week 12 of IFN treatment achieved poor treatment results. In the phase III trial of PEG-IFN-2α, which

Table 1 Cut-off values of HBsAg in the different phases of the natural course of HBV-infection

	Immunotolerance phase	Immunoactive phase	Inactive carrier phase
HBeAg positive CHB	- HBsAg >25,000 U/mL had >90%PPV for liver fibrosis <F1 [14] - HBsAg >100,000 U/mL suggestive for immune tolerance [13]	- lower HBsAg levels (<3.85 logIU/mL) are associated with fibrosis [15]	n.a.
HBeAg negative CHB	n.a.	HBsAg <1000 IU/mL linked to smaller risk for HCC [22]	HBsAg <1000 IU/mL and HBV DNA <2000 IU/mL corresponds to 87.9% PPV for inactive carrier phase [21]

HBsAg, hepatitis B surface antigen; PPV, positive predictive value; n.a., not available; HBV, hepatitis B virus; CHB, chronic hepatitis B; HCC, hepatocellular carcinoma

Table 2 Usefulness of HBsAg in the management of different settings of chronic HBV-infections

Therapy	Baseline	On treatment
IFN	Lower HBsAg associated with better treatment response [34]	HBeAg positive CHB
		HBsAg <300 IU/mL after 6 month, correlated with sustained virological response [36]
		HBsAg <1500 IU/mL at week 12 corresponds to 57% PPV for anti-HBe seroconversion and 17.6% HBsAg clearance [37]
		No decline of HBsAg at week 12 corresponds to 3-18% anti-HBe seroconversion [37,38]
		HBsAg >20,000 IU/mL associated with 100% NPV for anti-HBe seroconversion [31]
		HBeAg negative CHB
		HBsAg decline >0.5 log at week 12 lead to treatment response in 89% [40]
		No HBsAg decline (any decline) and <2 log decline of HBV DNA showed a NPV of 100% for nonresponse in genotype D patients [41]
NA	n.a.	HBsAg decline >1 log after 1 year corresponds with HBsAg loss [56]
		HBsAg decline >0.5 log 2 years after HBV DNA suppression correlated with HBsAg loss [52]
		HBsAg level <100 IU/mL predictive for sustained response 2 years after EOT [55]

HBsAg, hepatitis B surface antigen; IFN, interferon alpha; NA, nucleos(t)ide analogue; HBeAg, hepatitis B e antigen; HBV, hepatitis B virus; CHB, chronic hepatitis C; PPV, positive predictive value; NPV, negative predictive value; n.a., not available; EOT, end of treatment

included mostly Asians with predominantly genotype B and C, only 18% of patients without HBsAg decline achieved anti-HBe seroconversion [37]. Sonneveld *et al* reported even lower numbers of 3% anti-HBe seroconversion 6 months after treatment for patients without any HBsAg decline at week 12 of treatment, for a study population of mainly Caucasian descent with genotype A and D infection [38]. Again, this supports the importance of HBsAg quantification in relation to the HBV genotype. Overall, the NEPTUNE Study with 544 patients treated with PEG-IFN-2α also confirmed the importance of HBsAg decline. HBsAg >20,000 IU/mL was associated with 100% NPV for anti-HBe seroconversion [31]. Similar results were recently found by Sonneveld *et al* [39]. In conclusion, the NPV for anti-HBe seroconversion is high if the cut-offs for different genotypes are considered, but the PPV still warrants improvement.

HBsAg could also be useful in monitoring treatment response in HBeAg negative chronic hepatitis B (Table 2). At present, HBV DNA is mostly used as a marker for treatment response. HBV DNA may become negative during treatment, but still patients are at a considerable risk of relapse after the end of treatment [32]. In a small study in 48 patients with HBeAg negative chronic hepatitis B treated with PEG-IFN-2α for 48 weeks, there was a 89% chance of response if HBsAg declined >0.5 log at week 12 and >1 log IU/mL at week 24. In contrast, in patients who failed to achieve HBsAg decline, the chance of a response defined as undetectable serum HBV DNA (<70 copies/mL) 24 weeks after treatment cessation was only 10% [40]. Rijckborst *et al* studied both, HBV DNA and HBsAg levels, for treatment response. They showed that the absence of any HBsAg decline and no decline in HBV DNA >2 log were associated with no response to treatment [41]. In a validation study, they confirmed these results, but their study included mostly genotype D infected patients and the best performance was found for genotype D [42].

In conclusion, quantitative HBsAg improves the management of chronic hepatitis B with IFN treatment. Since IFN treatment should be considered as a finite treatment option, early stopping rules after 12 weeks of therapy can be applied with the use of quantitative HBsAg [4]. However, stopping rules differ between HBeAg positive and HBeAg negative patients and optimal cut-offs depend on the HBV genotype. Unfortunately, analysis of the HBV genotype is not reimbursed in most countries and thus is not available in the routine management. For general practice, we suggest to use futility rules independent of genotypes. Tolerability may be considered to decide if these rules should be applied very stringently. In HBeAg positive patients, IFN should be discontinued at week 12 if HBsAg does not decline more than the standard error of the applied test (usually 5-10%) or HBsAg is still >20,000 IU/mL. In HBeAg negative patients, no HBsAg decline and <2 log decline of HBV DNA at week 12 should result in IFN termination.

NA treatment

The aim of NA treatment is viral suppression during treatment. Therefore, treatment duration is, to begin with, unlimited. Long-term viral suppression improves liver histology and even reversion of cirrhosis over a treatment period of 5 years seems to be possible [43,44]. The long-term treatment with NAs also reduces the incidence of HCC, if potent NAs are used and no resistance occurs [45,46]. However, treatment with NAs is indefinite is most cases. The optimal endpoint and the only situation where NA treatment can be safely discontinued is HBsAg loss [4]. Discontinuation of NA treatment 12 months after anti-HBe seroconversion is also

possible but relapse is not infrequently observed [47]. As mentioned in the section about IFN treatment, the HBsAg decline in patients treated with NA is lower than in patients treated with IFN, though patients treated with NA have a marked drop in HBV DNA [48]. The reason for the lower decline in HBsAg in comparison to IFN treatment is probably due to the mechanism of action of NA treatment. NA treatment does affect the reverse transcription of pregenomic RNA, but does not affect the cccDNA and subgenomic RNA which have a translational activity associated with HBsAg levels [49].

As recently published by Chevaliez *et al*, it is estimated that with current NA treatment the median number of years needed to clear HBsAg is 52.2 years [50]. Another study by Zoutendijk *et al* predicted 36 years for HBsAg loss in HBeAg positive HBV infection and 39 years for HBeAg negative HBV infection [51]. There may be differences and some patients show some level of HBsAg decline while others will never show any drop of HBsAg level [52]. Some data suggest that HBsAg decline during NA is mainly found in HBeAg-positive patients and patients with genotype A [48]. It can be discussed that the stronger decline seen in HBeAg positive patients may be just due the natural course of infection as discussed above and could be observed also without NA treatment [52]. However, the strong effect on HBV DNA suppression with NA could potentially restore immune responses [53], which potentially results in HBsAg decline. This hypothesis is further supported by data that HBsAg decline was stronger in patients with higher ALT at baseline [51] or higher levels of interferon gamma-induced protein 10 (IP-10) at baseline, which indicates some level of immune response. IP-10 is a chemokine, which is secreted by different cell types in response to IFN-γ and may reflect the innate immune response against HBV [52]. Thus, HBsAg kinetics may be useful to monitor treatment responses during NA treatment as well. Wursthorn *et al* showed that a decline of HBsAg >1 log after 1 year of treatment with telbivudine in HBeAg positive patients is connected with a consecutive HBsAg loss. Supporting the aforementioned hypothesis, this was associated with markedly enhanced antiviral T cell reactivity [54]. Similar findings were observed in a study with 11 patients with HBeAg positive hepatitis B, who had been treated with telbivudine. A HBsAg level of <100 IU/mL was predictive for a sustained response 2 years after end of treatment [55]. For HBeAg negative patients HBsAg level <2 log IU/mL and a reduction >1 log from baseline were associated with a virological response, defined as HBV DNA <200 IU/mL 12 months post treatment [56]. In a German cohort of 95 HBeAg positive and HBeAg negative patients, HBsAg decline >0.5 log at 2 years after achieving HBV DNA suppression <100 IU/mL was correlated with HBsAg loss [52].

Because of the aforementioned long-term treatment with NAs, efforts have been made to identify possible stop points for NA treatment. Hadziyannis *et al* showed that in HBeAg negative infection with normalized ALT and suppressed HBV DNA after treatment with adefovir for 4-5 years, the withdrawal of antiviral therapy led to a sustained off-treatment HBV DNA suppression in 55% of patients, despite initial virological and biochemical relapse. Lower HBsAg levels at end of treatment were predictive for later HBsAg loss [57]. Again, this supports the idea that immune responses are important for the long-term control of HBV-infection. Some other small studies presented at recent meetings (AASLD and EASL) suggest that some patients with lower HBsAg levels are able to maintain HBV suppression after discontinuation of long-term NA therapy.

In summary, despite the fact that HBsAg decline is observed under NA treatment, the exact role is yet to be defined. At present, no cut-off values of HBsAg in NA treatment are established. Currently available data about the role of HBsAg in NA treatment is summarized in Table 2. Future studies are needed to investigate if withdrawal of long-term NA treatment is possible in patients with lower HBsAg levels.

Combination of NA and IFN treatment or switch strategies

Different approaches to combine IFN and NA have been performed to increase response to therapy. The first is an add-on therapy with IFN on an established treatment with NA. The second is a switch from NA to IFN.

Although the initial trials of PEG-IFN and lamivudine failed to show that the combination therapy was superior to IFN monotherapy [28,59], a retrospective analysis of HBeAg positive patients revealed a more significant decline of HBsAg levels on treatment with PEG-IFN plus lamivudine compared with PEG-IFN only [38].

Also other smaller studies showed some intriguing results combining IFN and NA. The combination of PEG-IFN-2β and adefovir proved to be effective in reducing intrahepatic cccDNA, HBV DNA in the serum and HBsAg. Four out of 26 patients achieved HBsAg loss [60]. Takkenberg and colleagues reported an astonishing 11% HBsAg loss in HBeAg positive and even 17% HBsAg loss in HBeAg negative patients two years after treatment with PEG-IFN and adefovir [34]. Thus, approaches to combine IFN and NA should be further evaluated. One concept could be adding IFN to a well-established NA treatment and monitoring HBsAg levels. Kittner *et al* observed that the add-on of PEG-IFN to NA induced HBsAg decline and even anti-HBs seroconversion in 2 out of 12 patients [61]. During recent meetings, several studies with add-on concepts have been presented at the EASL and AASLD. Wu Z *et al* showed promising results for the add-on concept in comparison with NA treatment: IFN add-on therapy led to HBsAg levels <10 IU/mL in 12 of 16 patients, who had 100-500 IU/mL before and HBsAg loss in 8 of 16 patients. In the control group, treated with NA solely, none of the patients achieved one of the 2 endpoints.

Another option could be a switch of NA to PEG-IFN, if finite treatment concepts have not been considered before. Ning Q *et al* compared the switch from NA treatment to IFN treatment for HBeAg positive patients, who had been on entecavir for at least 9 months before the switch. In comparison with the entecavir group, the IFN group showed significant HBsAg decline and achieved HBsAg loss in 9.3%, whereas the entecavir group had none after 48 weeks. Baseline HBsAg of <3000 IU/mL was associated with a positive response to therapy [62].

In summary, the combined therapy of IFN and NA can be effective, though the optimal concept for therapy is still unknown. HBsAg levels may help to investigate the use of this concept. Further studies are needed to define baseline cut-off values and on treatment levels to identify patients who have the highest profit of the combined approach and to predict therapy response.

Co-infection

HBV / HCV

The value of quantitative HBsAg in HBV/HCV co-infected patients is unknown. The virological patterns in HBV/HCV co-infection are widely divergent and have dynamic profiles. In most cases HCV is the dominating virus [63]. Treatment of HBV/HCV co-infection with anti-HCV treatment, i.e. PEG-IFN and ribavirin can result in high SVR rates for HCV [64]. However, eradication of HCV can result in reactivation of HBV [65].

We have initial data showing that HBsAg levels are lower in HBV/HCV patients with HCV dominance compared to active HBV monoinfection [66]. However, two distinct populations exist, one with HBsAg levels <1000 IU/mL and one with HBsAg levels >1000 IU/mL. Further studies need to evaluate if those patients with low HBsAg levels are protected from HBV reactivation after HCV is eliminated by antiviral treatment.

HBV / HIV

We have investigated HBsAg levels in 174 HBV/HIV co-infected patients. HBsAg levels were found to be correlated with the CD4 T cell count, reflecting the importance of immune responses for the control of HBV and the level of HBsAg. Patients with HIV/HBV co-infection had higher HBsAg levels compared to HBV mono-infected patients and patients on antiretroviral therapy had significantly lower levels of HBsAg than untreated patients [67]. A longitudinal follow-up showed that improved immune status corresponds with long-term decline of HBsAg levels in HBV/HIV co-infected patients [68]. A multicenter, prospective study in France with 143 antiretroviral-experienced HBV/HIV-co-infected patients examined the affect of tenofovir treatment on HBsAg and HBeAg. The median follow up was 30.3 months. HBsAg loss was noted in 4% of the study population. Baseline HBsAg levels <400 IU/mL were associated with HBsAg loss [69]. This may indicate stronger immune responses. These findings support the already mentioned fact that the baseline HBsAg level is important to predict further HBsAg loss.

HBV / HDV

Hepatitis delta requires the HBsAg for complete replication and transmission, therefore HDV infections only occurs as HBV co-infection or superinfection to an already existing HBV infection [70]. In most cases HDV suppresses HBV replication and, of note, HCV replication [71]. As of now, only IFN or PEG-IFN treatment has proved to achieve a virological response in HBV/HDV co-infected patients with sustained HDV RNA clearance in about 25% of patients as shown in the HIDIT-1 study [72]. However, Heidrich *et al* showed at EASL 2013 that the long-term follow up revealed that in 9 of 16 patients a relapse was noted [73]. Thus, HBsAg loss may be the only cure for HDV infection. For current treatment of the HBV/HDV co-infection with PEG-IFN, usually HDV RNA is monitored. Erhardt *et al* showed that patients with <3 log decline in HDV RNA after 24 weeks of treatment do not benefit from the treatment [74]. The role of HBsAg in HDV infection remains to be determined. Manesis *et al* suggested that monitoring of HBsAg levels in addition to HDV RNA in patients with chronic delta hepatitis provides more insight during treatment [75]. Ouzan *et al* reported 4 cases of hepatitis delta patients that had been treated with PEG-IFN until HBsAg loss [76]. HBsAg levels were used to guide therapy. This could be a concept for the treatment of hepatitis delta. However, time to achieve HBsAg loss on IFN therapy can be as long as 12 years as demonstrated by Lau *et al* in a similar case report [77].

Concluding remarks

HBsAg levels in the serum have been shown to reflect active intrahepatic cccDNA and to add additional value in treatment decisions, especially as an on treatment marker.

In the natural course, HBsAg helps to detect the different phases, with lower HBsAg levels associated with immune control by the host. Furthermore, lower HBsAg levels are connected with a lower risk for HCC development.

In IFN treatment, HBsAg levels can help to identify patients early who fail to respond to treatment. In NA treatment, HBsAg drop is associated with future HBsAg loss and may help identify patients in whom NA treatment can be stopped. HBsAg levels may also be beneficial in future treatment concepts to increase HBsAg loss. To date, these concepts include combination therapies of NA and IFN. Pitfalls in the use of HBsAg are the variety of HBsAg levels in dependence of HBV genotypes. HBV mutations such as pre-S mutations may also account for different levels of HBsAg. For the treatment of HBV co-infections, the role of HBsAg remains unclear due to fewer studies with an overall smaller number of patients. However, HBsAg levels may help to further comprehend the viral interactions and improve the management of HBV co-infections.

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