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# Quantitative hepatitis B core antibody and quantitative hepatitis B surface antigen: Novel viral biomarkers for chronic hepatitis B management



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**Abstract**

The management of hepatitis B virus (HBV) infection now involves regular and appropriate monitoring of viral activity, disease progression, and treatment response. Traditional HBV infection biomarkers are limited in their ability to predict clinical outcomes or therapeutic effectiveness. Quantitation of HBV core antibodies (qAnti-HBc) is a novel non-invasive biomarker that may help with a variety of diagnostic issues. It was shown to correlate strongly with infection stages, hepatic inflammation and fibrosis, chronic infection exacerbations, and the presence of occult infection. Furthermore, qAnti-HBc levels were shown to be predictive of spontaneous or treatment-induced HBeAg and HBsAg seroclearance, relapse after medication termination, re-infection following liver transplantation, and viral reactivation in the presence of immunosuppression. qAnti-HBc, on the other hand, cannot be relied on as a single diagnostic test to address all problems, and its diagnostic and prognostic potential may be greatly increased when paired with qHBsAg. Commercial qAnti-HBc diagnostic kits are currently not widely available. Because many methodologies are only semi-quantitative, comparing data from various studies and defining universal

cut-off values remains difficult. This review focuses on the clinical utility of qAnti-HBc and qHBsAg in chronic hepatitis B management.

**Key Words:** Quantitative hepatitis B core antibody; Quantitative hepatitis B surface antigen; Chronic hepatitis B management; Novels viral biomarkers

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**Core Tip:** It is possible to employ a quantitative hepatitis B virus (HBV) core antibody (qAnti-HBc) level as a predictor of therapeutic response, recurrence after hepatitis B surface antigen (HBsAg) loss, and HBsAg seroclearance. Just like other newly identified biomarkers, qAnti-HBc is not a stand-alone diagnostic test that can solve every issue. The information that is now available indicates that it may have a much higher diagnostic and prognostic effectiveness when combined with quantitative HBsAg. Further research involving larger and more variable patients is required to assess the actual usefulness of these biomarkers.

## INTRODUCTION

Hepatitis B virus (HBV) is a chronic infection that affects 250 million people globally, 10% to 15% of whom develop chronic liver disorders such as chronic hepatitis B (CHB), cirrhosis, and hepatocellular carcinoma (HCC). The incidence of cirrhosis and HCC can now be greatly decreased by attaining both HBV DNA undetectable and hepatitis B surface antigen (HBsAg) clearance with or without hepatitis B surface antibody (HBsAb) appearance, known as functional cure[1-3]. However, the effectiveness of currently available medications, such as nucleoside or nucleotide analogs (NAs) and interferon (IFN), is still insufficient when taken as a single medication. The likelihood of

prolonged HBsAg loss or seroconversion in CHB patients has significantly increased because of newly discovered novel techniques that switch to or add on pegylated-interferon-alpha (PEG-IFN-α) to ongoing NA therapy[4-7]. After 48–96 wk of medication, the loss rate in patients with quantitative HBsAg (qHBsAg) < 1500 IU/mL, with or without early on-treatment HBsAg reduction, can range from 22% to 58%. Moreover, around 40% to 80% of these patients still test positive for HBsAg even after experiencing PEG-IFN-α side effects[8-11]. Thus, it is essential to develop novel markers or techniques to accurately pinpoint the patients who have the best chance of achieving HBsAg clearance while receiving PEG-IFN-α-based treatment. Two innovative indicators for patients with CHB are the quantitative anti-hepatitis B core antibody (qAnti-HBc) and qHBsAg. Research has demonstrated that in CHB patients, qAnti-HBc levels are much greater during the immune clearance and reactivation stages of natural history than they are during the immunological tolerance and low replication phases[12-14]. In patients who are new to treatment, there is a close correlation between the qAnti-HBc levels and serum ALT levels, as well as the degree of hepatic inflammation. Over the course of NA medication, there was a steady decrease in these levels[15,16]. With regard to qHBsAg, it is utilized to track antiviral medication response, forecast recurrence following treatment termination, and calculate the risk of HCC[17-19]. It is yet unknown, nevertheless, if qAnti-HBc and qHBsAg levels in NA-suppressed CHB patients undergoing NA therapy alone or in conjunction with PEG-IFN-α can forecast HBsAg clearance. The purpose of this review was to evaluate the kinetics of serum qAnti-HBc and qHBsAg levels in CHB patients who were treated with NAs alone or with PEG-IFN-α as an adjuvant to continuous NA therapy. Additionally, the significance of these two new biomarkers in the context of treatment response prediction, the occurrence of recurrence subsequent to the loss of HBsAg, and the phenomenon of HBsAg seroclearance are considered.

## LIFE CYCLE OF HBV

The HBV belongs to the family *Hepadnaviridae* and is an enveloped DNA virus. The antigen for HBV, which is now known as surface antigen but was once named "Australia antigen" according to research published in 1965 by Blumberg and colleagues, was initially found in an Australian aborigine[20]. Independent reports of this antigen's existence in hepatitis patients date back to 1968 from Prince, Okochi, and Murakami[21,22]. Dane and colleagues used an electron microscope to visualize the viral particles in 1970[23]. In the blood of individuals who have contacted the infection, at least three different types of HBV particles have been identified: filament structures of varying length with a diameter of 22 nm, spherical structures measuring 42 nm, and those with a diameter of 22 nm. A nucleocapsid made up of the viral genome DNA, viral polymerase (Pol), and the hepatitis B core protein (HBc) is encased in a lipid membrane containing three different viral surface antigens (HBs): large (L-HBs), middle (M-HBs), and small (S-HBs). These 42 nm particles are also known as Dane particles. Subviral particles (SVPs), which are non-infectious due to their absence of nucleocapsid, are among the 22 nm particles seen in patient serum, which are significantly more prevalent. Moreover, various non-infectious particles, such as envelope-less particles (naked nucleocapsids), those carrying viral RNA, and enclosed particles without a viral genome, are now known to be created during infection[24].

The relaxed-circular DNA (rcDNA) that makes up the HBV genome is around 3.2 kb long and has an incomplete plus strand and a full minus strand. Four overlapping open reading frames (ORFs), C, P, S, and X, are encoded by the viral genome and are the source of functional viral proteins. From C, HBc and related proteins like E antigen (HBe) and 22-kDa pre-core protein (p22cr) are produced; from P, Pol is produced; from S, three different types of surface antigens are produced: L-HBs, M-HBs, and S-HBs; and from X, HBV X protein (HBx) is produced. In infected cells, rcDNA is transformed into covalently closed circular DNA (cccDNA). CccDNA then generates HBV RNAs,

namely 3.5 kb, 2.4 kb, 2.1 kb, and 0.7 kb, which are transcribed from various promoters in the HBV. The protein product from C and P is generated by a 3.5-kb RNA; L-HBs are translated by a 2.4-kb RNA, and M-HBs and S-HBs are synthesized by a 2.1-Kb RNA; HBx is produced by a 0.7-kb RNA. The resulting HBc protein product first combines to form a dimer *via* its N-terminal domain. Subsequently, it self-assembles to form an icosahedral capsid made up of 90 or 120 dimers, which includes the 3.5-kb viral pregenomic RNA (pgRNA) linked to Pol (more on this below). The 3.5-kb preC mRNA, which has a prolonged 5' upstream region of the C gene and is translated into HBe, is then cleaved at the C-terminus of the resulting protein. The largest HBV protein, Pol, is made up of four domains, each of which has a distinct enzymatic function: (1) the spacer domain, whose function is unclear; (2) the terminal protein (TP) domain, which is vital for binding to pgRNA and acts as a protein primer to start minus-strand DNA synthesis; (3) the reverse transcriptase (RT) domain, which has DNA elongation activity for both reverse transcription and DNA-dependent DNA polymerization; and (4) the ribonuclease H (RNaseH) domain, which digests pgRNA following reverse transcription. The C-terminal S region of all three HBs proteins is identical. Furthermore, an expanded area at the N-terminus of the S region, known as the preS2 region, is carried by M-HBs and L-HBs. At the N-terminus of the preS2 and S sections, L-HBs also have an extension (preS1 region) that is crucial for receptor binding during viral entry. The multifunctional protein HBx contributes to the development of HCC associated with HBV and stimulates the creation of viruses at several stages, including transcription and replication. By using these viral proteins in conjunction with host-derived stimuli, HBV multiplies in host cells[25,26].

## ORIGIN AND DEGRADATION OF CIRCULATED HBsAg

Serum HBsAg is made up of cccDNA and integrated HBV DNA. The former can exist in either a compacted, transcriptionally inactive state or a relaxed, transcriptionally active

state. Infectious HBV particles are generated and released into the blood, as are noninfectious SVPs[27]. The latter is HBV DNA that has been incorporated into various regions of the hepatocyte chromosome. Because the integrated DNA lacks a typical circular shape, it can only produce S- and M-HBsAg, and the spherical SVPs released into the blood cannot synthesize pregenomic RNA (pgRNA) or other viral proteins[28,29]. SVPs contain 99.99% of the HBsAg in the blood. The majority of blood HBsAg in CHB patients with a full virological response or zero hepatitis B e-antigen (HBeAg) derives from integrated HBV DNA rather than cccDNA[30,31]. HBV DNA that is transcriptionally active and integrated is present throughout the liver and generates broad HBsAg independent of HBV replication. Infected hepatocytes control HBsAg secretion by a number of breakdown mechanisms, including endoplasmic reticulum-mediated proteolysis and autophagy. Furthermore, the degradation of M- and L-HBsAg is aided by the unique proteolytic mechanisms of the proteasome, ubiquitin, and proteome-independent mechanisms[32-34]. When hepatocytes are infected, the activity of the degradation pathways increases, demonstrating that HBsAg renewal is implicated in the synthesis of SVP and virus.

## PATTERN OF HBsAg SEROCLEARANCE

With a yearly incidence of about 1%, spontaneous HBsAg seroclearance is an uncommon occurrence[35]. The cumulative rates of spontaneous HBsAg seroclearance after 10 and 25 years, respectively, were 8.1% and 44.7%, according to research with 1076 CHB patients[36]. The crude incidence rate of HBsAg loss was 1.6% per 100 person-years, according to a prospective follow-up of 1240 patients with negative HBeAg who had not received treatment for 5.5 years. More significant correlations between HBsAg seroclearance and quantitative HBsAg levels, non-Asian race, older age, inactive HBsAg carriers, HBV genotype A, and lower HBV DNA were also discovered[37]. Instead of directly interacting with cccDNA, NAs only inhibit HBV DNA. Once they are

phosphorylated, they work as antimetabolites by being similar enough to nucleotides to be incorporated into growing DNA strands; but they act as chain terminators and stop viral DNA polymerase. As a result, NAs find it extremely challenging to stop the synthesis of HBV particles and their antigens. In 26614 person-years of entecavir (ETV) or tenofovir dipivoxil (TDF), the 10-year cumulative loss rate of HBsAg was just 2%, according to a new large multicenter cohort study that included 4769 CHB patients[38]. HBsAg loss may result after discontinuing NAs in accordance with the recommendations' withdrawal criteria, notwithstanding the possibility of virological recurrence, clinical relapse, or worsening of the liver disease[39,40].

## DURABILITY AND CLINICAL OUTCOMES OF HBsAg SEROCLEARANCE

Without focusing on the removal of integrated HBV DNA and cccDNA, functional cure is more closely associated with the restoration of liver function, particularly the specific immune function against HBV, *via* the maximal long-term suppression of HBV replication. There is still a significant difference between a functional cure and a full recovery. The length of functional cure following medication withdrawal and the improvement of long-term outcomes are also critical indicators of complete cure, in addition to the detection of integrated HBV DNA and cccDNA. In a retrospective cohort study, 4568 CHB patients with HBsAg clearance were included; of them, 793 had undergone NA treatment and 60 underwent interferon (IFN) therapy. Fifty-four individuals (2.9%), including 49 who had spontaneous HBsAg clearance and five males over 50 receiving NA treatment, developed liver cancer over a median follow-up of 3.4 years. At one, three, and five years, the cumulative incidence rates of HCC were 0.9%, 1.3%, and 1.5%, respectively. Within five years, no patient receiving PEG-IFN-α developed liver cancer. Two independent predictors of the likelihood of HCC in patients with HBsAg clearance were sex and age[41]. In addition, 7124 CHB patients with HBsAg loss were also evaluated. There were 1207 cases of NA-induced clearance and

5917 cases of spontaneous clearance. There was no discernible difference in the incidence of HCC at 1.6% and 1.3%, respectively, after an average follow-up of 4.3 years[41]. Additionally, to determine the HBsAg status of each end point and the composite end point, Anderson *et al*[42] conducted a systematic review and meta-analysis of rate ratios (RR) using a random-effects model independently. They also reported the incidence of liver decompensation, liver transplantation, HCC, and all-cause mortality. 188316 patients with chronic HBV infection and 1486081 person-years (PY) of follow-up evaluation were included in the 28 studies whose data they evaluated; 26 of the studies contained data on HCC, 7 on liver decompensation, and 13 on liver transplantation and/or death. For both the HBsAg seroclearance group and the HBsAg-persistent group, the composite event rates were 2.45/1000 PY and 0.19/1000 PY, respectively. In the HBsAg seroclearance group, the pooled relative risks (RRs) for liver decompensation, HCC, liver transplantation, and/or mortality and the composite endpoint were 0.28, 0.30, and 0.22, respectively. Within subgroups of various research or patient characteristics, no variations in RR estimations were noted. They discovered a substantial correlation between improved patient outcomes and seroclearance of HBsAg. It is uncertain if individuals with CHB who acquired HBsAg seroclearance naturally or as a result of anti-viral treatment had similar clinical outcomes. Several studies examined the risk of HCC, hepatic decompensation, and recurrence of HBsAg positive in CHB patients who cleared HBsAg spontaneously or after anti-viral treatment with NAs alone, IFN-α alone, or IFN-α combination with NAs. Table 1 summarizes these clinical results[43-62].

Recently, Vittal *et al*[63] also conducted a meta-analysis comprised of 57 papers throughout the globe which recruited 2 prospective population-based studies, 22 prospective cohort studies, and 33 retrospective cohort studies. Out of the 258744 HBsAg-positive patients in total, 63270 (24.4%) had HBsAg loss. There were ten studies with patients who lost HBsAg spontaneously, twelve with patients who lost HBsAg as a

result of therapy, twenty-two with patients who lost HBsAg as a result of both treatment and spontaneous means, and thirteen without reporting the method of HBsAg loss. They discovered that HBsAg loss is linked to a lower likelihood of developing HCC as well as other significant clinical outcomes such as hepatic decompensation, liver cirrhosis, and death from all causes. The use of HBsAg seroclearance as the major end point of trials for individuals with persistent HBV infection is supported by the consistent outcomes across various study types and patient subpopulations studied. Additionally, Song *et al*[64] also conducted a meta-analysis to assess the HCC incidence and durability of HBsAg seroclearance following therapy discontinuation. They used a random-effects model to analyze the data. In the end, 38 studies and 43924 patients were included. The pooled recurrence rate of 6.19% demonstrated the durability of HBsAg seroclearance. Recurrence rates following various seroclearance techniques, as well as rates across recurrence kinds and geographical areas, did not differ significantly. The rate of recurrence was considerably lower after anti-HBs seroconversion. Compared to HBsAg-positive patients, those who had HBsAg seroclearance had a noticeably decreased incidence of HCC. Following HBsAg seroclearance, the pooled incidence of HCC was 1.88%; among patients without cirrhosis at baseline, this rate was 0.76%. Additionally, Liu *et al*[65] conducted a meta-analysis and systematic review of 28 studies encompassing 34952 individuals who had HBsAg seroclearance. Despite HBsAg seroclearance, the aggregate pooled percentage indicated that 2.29% of CHB patients will develop HCC. The pooled percentage of HCC development in HBsAg seroclearance patients without cirrhosis or HCV co-infection was 1.55%. Furthermore, individuals who had cirrhosis or were older than 50 years at the time of HBsAg seroclearance had a markedly increased chance of developing HCC. However, as compared to chronically positive HBsAg, seroclearance of HBsAg was substantially linked to a lower risk for HCC. They concluded that while HBsAg seroclearance can dramatically lower the risk for HCC, certain CHB patients may still

develop HCC following HBsAg seroclearance. Because HBsAg loss is linked to a more persistent reduction of viremia, the physicians favored using it as the surrogate endpoint of therapy rather than HBV DNA.

**qHBsAg**

qHBsAg tests have been used for the detection of hepatitis B infection since the 1970s. The expression of HBsAg occurs at an early stage in the viral life cycle. There has been a recent surge in interest in the measurement of HBsAg and its potential as a predictor of therapy results. Various commercially available qHBsAg assays have been developed, including Elecsys HBsAg II Quant by Roche Diagnostics and ARCHITECT QT by Abbott Laboratories. Both options are offered at a reasonable price, have undergone standardization procedures, and use an automated two-step chemiluminescence method. In the ARCHITECT QT test, the HBsAg, paramagnetic microparticles, and acridinium-labeled anti-HBs conjugate are combined in a sandwich configuration. The chemiluminescent outcomes are assessed using relative light units and then converted into an HBsAg quantification. The Elecsys HBsAg II Quant test employs a specific technique in which HBsAg, streptavidin-coated microparticles, and biotin-ruthenium-labeled monoclonal antibodies are combined in a sandwich configuration. The mixture is subjected to a provided voltage while it is drawn into a measuring cell, resulting in the generation of ruthenium chemiluminescence. This chemiluminescence may then be quantified using a photomultiplier. The automated dilution feature has the potential to elevate the detection limit of both assays to levels above 50000 IU/mL. The dynamic ranges of the tests are 0.05–250 IU/mL for the ARCHITECT QT assay and 0.05–130 IU/mL for the Elecsys HBsAg II Quant assay. The HISCL HBsAg test has been recently developed for the quantification of HBsAg using the chemiluminescence enzyme immunoassay (CLEIA) technique, using the CDP-Star® chemiluminescent substrate. The biotinylated monoclonal antibodies targeting HBsAg derived from mice exhibit a

particular reaction with HBsAg present in the sample. These antibodies subsequently attach themselves to streptavidin-coated magnetic particles. Following the process of bound/free (B/F) separation, the monoclonal antibodies (mouse) tagged with alkaline phosphatase (ALP) exhibit specific binding to the HBs antigen on a microparticle. Subsequently, the ALP present on the membrane protein (MP) cleaves the CDP-Star® substrate, resulting in the formation of an energetically excited intermediate that gives rise to a luminous signal. The assay samples were evaluated at their original concentration using an analytical measurement range spanning from 0.03 to 2500 IU/mL, without the need for any preliminary dilution. Nevertheless, the system is equipped with an automatic dilution feature to address instances when HBsAg concentrations are very high. Additionally, a Lumipulse HBsAg-HQ test was created to assess HBsAg concentrations. The measurement of HBsAg was conducted using the two-step sandwich assay technique, using a fully automated chemiluminescent enzyme immunoassay system known as Lumipulse G1200, manufactured by Fujirebio, Inc. The samples underwent pretreatment using a solution containing a surfactant in order to disrupt HBV particles. This process aimed to detach HBsAg from HBsAg-anti-HBs complexes and denature epitopes into a linear form. The detection of linearized HBsAg was accomplished by using two monoclonal antibodies that target exterior structural areas, namely determinant "a," as well as an internal epitope that served as a capture reagent. Additionally, two monoclonal antibodies linked to ALP were used as detectors. During the test methods, 100 µL of blood serum and/or plasma samples, together with 20 µL of pretreatment solution, were subjected to incubation with monoclonal antibodies that bind to ferrite microparticles. This incubation took place at a temperature of 37 °C for a duration of 10 min. Following the completion of the automated washing process, a volume of 250 µL of ALP-labeled antibodies was introduced and then incubated at a temperature of 37°C for a duration of 10 min. Following the washing procedure, a volume of 200 µL of the substrate solution,

AMPPD, was introduced and then incubated at a temperature of 37 °C for a duration of 5 min. The measurement of chemiluminescence intensity was conducted in order to determine the concentration of HBsAg by comparing it to a standard curve. The assay included a range of HBsAg values from 0.005-150 IU/mL. In cases where the concentration exceeded this range, retesting was conducted using a 100, 200 or 1000-fold dilution of the samples. The development of iTACT-HBsAg occurred in 2016, using monoclonal-antibody-coated ferrite particles as the solid phase *via* the coupling approach with chemical linkers. The particle solution was prepared by suspending ferrite particles coated with antibodies in a diluent specifically designed for particle suspension. The detection reagent consisted of ALP-linked monoclonal antibodies that were produced using the hinge approach and then purified by chromatography using a HiLoad 16/600 Superdex 200 pg column. The preparation of the conjugate solution included the process of diluting the ALP conjugate in the conjugate diluent. The iTACT-HBsAg test was conducted using the LUMIPULSE PRESTO II, an automated CLEIA instrument manufactured by Fujirebio. Fifty µL of samples were subjected to incubation with a 30 µL acidic pretreatment solution for a duration of 6.5 min at a temperature of 37 °C. The samples that had been pre-treated were thereafter combined with a 50-µL solution containing particles and incubated for a duration of 8 min at a temperature of 37 °C. Following the washing of the ferrite particles with Lumipulse Presto washing buffer, an incubation period of 8 min at 37 °C was conducted with the conjugate solution. Subsequently, the relative luminous intensity was assessed by incubating the particles for 4 min at 37 °C with 200 µL of substrate solution (Table 2)[66-71].

## qHBsAg AND FUNCTIONAL CURE

Chtourou *et al*[72] conducted a study to assess the diagnostic utility of qHBsAg in detecting HBV functional cure among 174 Tunisian patients with HBeAg-negative CHB infection over a 1-year prospective follow-up period. During the follow-up period,

it was found that 21.6% of patients with a low initial viral load (< 2000 IU/mL) and 19.5% of patients with an intermediate viral load (2000–20000 IU/mL) experienced an increase in their HBV DNA levels above 2000 and 20000 IU/mL, respectively. Notably, significant fluctuations in viral load were observed in 61.1% of patients at 6-month intervals. Among the 174 patients, 89 (51.1%) belonged to inactive carriers, 33 (19%) to patients with negative HBeAg CHB, and 52 (29.9%) to patients with an indeterminate state. Out of the 14 patients who underwent a liver biopsy, seven exhibited moderate-to-severe liver disease. The combination of HBV DNA <2 000 IU/mL and qHBsAg < 832 IU/mL effectively ruled out CHB in 98.4% of cases. Furthermore, a qHBsAg cutoff point of < 100 IU/mL, coupled with an annual decline of >0.5 Log 10 IU/mL, proved to be a reliable predictive marker for achieving a functional cure in CHB patients. It was discovered that individuals with HBeAg-negative chronic HBV infection, namely those with genotype D, had significant short-term changes in HBV DNA levels. Therefore, by using the qHBsAg cutoff value of 832 in conjunction with the HBV DNA cutoff value of 2000, it becomes feasible to effectively rule out CHB in the majority of patients. The authors proposed doing an initial assessment of patients by measuring ALT, HBV DNA, and qHBsAg levels. In instances where values fall below the established thresholds, it may be appropriate to conduct further monitoring *via* the use of ALT and qHBsAg tests only.

Coffin *et al*[73] conducted a retrospective review of clinical outcomes in a broad community of patients with CHB who obtained functional cure, defined as HBsAg loss. The study compared these patients to people with different levels of HBsAg using the quantitative HBsAg assay. A total of 844 individuals diagnosed with CHB were included in the study. Among these patients, 237 (28.0%) tested negative for HBsAg, while 190 (22.5%) had a qHBsAg level ranging from 1 to 100 IU/mL. Additionally, 91 (10.8%) patients had a qHBsAg level between 100 and 500 IU/mL, 54 (6.4%) had a qHBsAg level between 500 and 1000 IU/mL, and 272 (32.2%) had a qHBsAg level

beyond 1000 IU/mL. In general, a total of 682 individuals, accounting for 80% of the sample, had documented HBeAg status at the final follow-up. The predominant proportion, namely 87.0%, exhibited a negative HBeAg status. Furthermore, it is worth noting that 54% of the participants in the study, namely 461 out of 844 individuals, had previously had antiviral medication. The patients who received treatment exhibited a reduced likelihood of developing cirrhosis or HCC in comparison to the individuals who did not get treatment. The HBsAg-loss group had a lower prevalence of cirrhosis in comparison to the HBsAg-positive group, with rates of 5.7% and 10.9%, respectively. Additionally, the HBsAg-loss group showed a lower incidence of HCC at 0.9% compared to the HBsAg-positive group at 6.2%. The predominant group of patients who had HBsAg loss consisted of individuals who had not received treatment, were negative for HBeAg, exhibited undetectable levels of HBV DNA, and demonstrated normal liver enzyme levels. The presence of low-level qHBsAg (< 100 IU/mL) was seen in individuals who had HBsAg loss, as determined by testing conducted within one year following HBsAg seroclearance. The investigators concluded that individuals with CHB who had antiviral medication and experienced a reduction of HBsAg had a reduced likelihood of developing cirrhosis and HCC. There was no observed correlation between the amount of qHBsAg and the presence of hepatic fibrosis. Individuals who successfully attained HBsAg loss had low-level qHBsAg during a period of one year following seroclearance.

In order to compare the efficacy of two novel assays — iTACT-hepatitis B surface antigen (iTACT-HBsAg) and iTACT-hepatitis B core-related antigen (iTACT-HBcrAg) — with 120 HBsAg-negative and anti-HBc-positive individuals, Wong *et al*[74] conducted a study in which 556 serial sera were collected from 96 CHB patients who had HBsAg seroclearance as confirmed by standard assays. Sixty seronegative people who tested negative for HBsAg, antiHBc, and anti-HBs made up the control group. They discovered that 154/418 (36.8%) of the samples obtained following seroclearance

could detected HBsAg. In samples taken before and after seroclearance, HBcrAg was found in 78.3% and 65.9% of the samples, respectively. Following seroclearance, the detectability rates of HBsAg and HBcrAg gradually dropped. Ten years following seroclearance, detectable HBsAg and HBcrAg were still present in 20.4% and 64.5% of the patients, respectively. 66 individuals (71%) had HBsAg and/or HBcrAg that could be detected. Eleven (9.2%) and four (3.3%) of the 120 HBsAg-negative and anti-HBc-positive people showed detectable HBsAg and HBcrAg, respectively. In the controls, HBsAg and HBcrAg were not detected. They concluded that the iTACT tests found that >70% of patients had low levels of HBsAg and/or HBcrAg even ten years after seroclearance, indicating that CHB patients who had seroclearance may still have low levels of HBV protein expression. Research on the clinical implications of detectable viral proteins following seroclearance in relation to the progression of the disease and HBV reactivation is warranted.

Seroclearance of HBsAg remains infrequent in CHB infections. ALT rise during acute flares of CHB (AFOCHB) indicates an increasing immune response aimed at clearing the virus. Hui *et al*[75] postulated that there is a correlation between severe AFOCHB and a more significant decrease in qHBsAg levels, as well as a higher rate of HBsAg seroclearance. A total of 75 patients diagnosed with severe AFOCHB and having ALT levels 10 times higher than the upper limit of normal (ULN) were selected. These patients were then matched with a control group based on age and sex in a 1:2 ratio. QHBsAg levels were assessed throughout the flare and yearly until the final follow-up. They discovered that the rate of HBsAg seroclearance was greater in the severe AFOCHB group compared to the control group (11.8% *vs* 5.0%, $P = 0.04$), even though the former group had a somewhat higher baseline median qHBsAg (3127 IU/mL *vs* 1178 IU/mL, $P = 0.076$). In comparison to the control group, the severe AFOCHB group had a higher yearly decrease in qHBsAg levels (-242.4 IU/mL/year vs. -47.3 IU/mL/year, $P = 0.002$). HBsAg seroclearance was independently linked with

increasing age ($P$ = 0.049), decreased baseline qHBsAg ($P$ = 0.002), and severe AFOCHB ($P$ = 0.014). Nevertheless, the overall incidence of HCC was much greater in the severe AFOCHB group compared to the control group (15.8% *vs* 1.9%, $P$ < 0.001). Caviglia *et al*[76] examined the clinical significance of qHBsAg in a real-world group of CHB patients receiving NAs therapy in a specialized medical facility in North-West Italy. A retrospective enrollment was conducted for a total of 101 patients with CHB (86 of whom were HBeAg-negative) who were having treatment with NAs. The level of HBsAg was assessed at four time points: baseline (T0), 6 months (T1), 12 months (T2), and the final follow-up (FU). The median follow-up period was 5.5 years, ranging from 3.2 to 8.3 years. After the follow-up period, 11 patients had a loss of HBsAg, resulting in an annual incidence rate of 1.8%. The initial levels of HBsAg were substantially higher in patients who did not experience an HBsAg decrease compared to those who had a functional cure (3.46 *vs* 1.11 Log IU/mL, $P$ < 0.001). The reduction in HBsAg from T0 to T2 was substantially different between the two groups of patients. The first group had a decline of 0.05, ranging from -0.04 to 0.13 Log IU/mL, while the second group had a decline of 0.38, ranging from 0.11 to 0.80 Log IU/mL. This difference was statistically significant, with a $P$-value of 0.002. The stratified cross-validation study demonstrated that the combination of baseline HBsAg and ΔHBsAg T0–T2 had a very high accuracy in predicting HBsAg loss, with a C statistic of 0.966. These findings confirm the effectiveness of qHBsAg in predicting HBsAg seroclearance in Caucasian CHB patients undergoing antiviral treatment.

Participants in the retrospective analysis were CHB patients who had not previously had entecavir medication but who had received at least two years of consecutive treatment in order to determine the usefulness of qHBsAg kinetics during long-term entecavir treatment. Abdominal sonography, liver biochemistry, and HBV DNA were used for patient follow-up at 3- to 6-month intervals. Patients who tested positive for HBeAg had their levels checked every three to six months until the findings stopped

coming back positive. We measured serum qHBsAg levels at baseline, one year, and five years. Biopsies of the liver, imaging tests, or the presence of portal hypertension in the patient's symptoms led to the diagnosis of liver cirrhosis. Histological analysis or research using dynamic imaging techniques confirmed the presence of hepatocellular cancer. Patients who tested positive for HBeAg had an earlier virological response, biochemical response, and HBeAg seroconversion when their baseline qHBsAg levels were lower; patients who tested negative for HBeAg but did not have cirrhosis had an earlier virological response. Despite the gradual reduction in qHBsAg levels seen following long-term entecavir therapy, patients with higher baseline HBsAg levels and those who tested positive for HBeAg but did not have cirrhosis had the fastest qHBsAg decline rates in the first year. Rapid HBsAg fall was not associated with better clinical outcomes; rather, it was dependent on lower HBsAg levels to begin with. Results showed that baseline HBsAg is a good predictor of response to therapy. When trying to make sense of qHBsAg changes, it's important to take both the levels and the rates of reduction into account with the patient's illness condition[77]. The objective of the study conducted by Ma *et al*[78] was to determine the factors that might predict HBsAg seroconversion in patients with CHB who are undergoing antiviral medication. They investigated the blood levels of quantitative pg-RNA, HBcrAg, and HBsAg as potential predictors. A cohort of 335 patients on antiviral treatment was included in the study. Out of them, only 23 patients achieved seroconversion for HBsAg. Additionally, a random selection of 138 individuals without seroconversion of HBsAg was made from the remaining 312 patients. The samples from a total of 161 patients were tested at various time intervals. The reduction in titers of pg-RNA, HBcrAg, and HBsAg from baseline to 6 months and baseline to 12 months was denoted as Δpg-RNA, ΔHBcrAg, and ΔHBsAg, respectively. Subsequently, Δpg-RNA, ΔHBcrAg, and ΔHBsAg were used as predictors for HBsAg seroconversion. They discovered that a total of 6.9% of the patients achieved seroconversion of HBsAg after a median duration of 3.61 years of

therapy. The receiver operating characteristic (ROC) analysis was used to predict the seroconversion of HBsAg. At 6 months, a ΔHBsAg of 0.64 Log10 IU/mL yielded an area under the curve (AUC) of 0.886, indicating a strong predictive ability. Similarly, at 12 mo, a ΔHBsAg of 1.45 Log10 IU/mL resulted in an AUC of 0.939, indicating a high level of predictive accuracy. These values were shown to have the highest Youden's index, indicating optimal predictive performance. The present study used the Kaplan-Meier approach to compare the frequencies of HBcrAg conversion between two groups of patients: 23 individuals who had HBsAg conversion and 138 individuals who did not. The analysis revealed a statistically significant difference between the two groups at the point of antiviral cessation. Based on the findings, ΔHBsAg has the potential to serve as a useful indicator for identifying patients who are likely to achieve seroconversion *via* adherence to antiviral medication. This is of significant importance in attaining the objective of a functional cure, or perhaps a clinical cure.

To estimate the length of treatment required to produce a functional cure, Cho *et al*[79] performed a study to look at the on-treatment dynamics of qHBsAg during entecavir therapy. A linear mixed model was used to evaluate the kinetics of qHBsAg reduction in 410 individuals from a cohort of 1009 CHB treatment-naïve patients initiated on entecavir. Based on baseline liver cirrhosis, HBeAg positivity, and HBeAg seroclearance, the variation in the kinetics of qHBsAg was ascertained. With a median age of 48 years, 213 (52.0%) and 217 (66.1%) male patients were among the 410 patients. Over a median follow-up of 53.5 months, there was a gradual but steady decline in the qHBsAg level. For HBeAg (+) and HBeAg (-) patients, the anticipated log qHBsAg values as a function of time during entecavir therapy were 3.4773 - 0.0039 × months and 3.1853 - 0.0036 × Months, respectively. For HBeAg-positive patients in this study, the expected time to clearance quantitative HBsAg was more than 74.1 years, whereas for HBeAg-negative individuals, it was 73.5 years. Without considering the occurrence of liver cirrhosis or HBeAg seroclearance, the estimated period to attain a functional cure

is a lifetime. According to mathematical modeling based on a long-term follow-up of patients with CHB using entecavir, clearing HBsAg takes decades of therapy. Therefore, to obtain a functional cure in individuals treated with entecavir, lifetime medication is required. For patients who test positive for HBsAg, HBeAg seroconversion represents a significant spontaneous shift and therapy endpoint. It is also a requirement for HBsAg loss or functional cure. In a retrospective analysis, Wang *et al*[80] used serum qHBsAg and quantitative hepatitis B core-related antigen (qHBcrAg) to predict seroconversion in HBeAg-positive patients receiving NA treatment. A retrospective analysis was done on 118 HBeAg-positive individuals (genotypes A–G) at various time points. ROC analysis was used to estimate the predictive potential of the on-treatment levels of qHBsAg and qHBcrAg, with cut-off values set by maximizing Youden's index. A median of 39 months of therapy resulted in HBeAg seroconversion in around 36.4% of patients. Regarding the HBV DNA treatment kinetics, there were differences in qHBsAg and qHBcrAg between HBeAg seroconverters and non-seroconverters. For HBeAg seroconversion, a combination of qHBsAg and qHBcrAg had the highest predictive value: at 6 months, a combination of 3.9 Log10 IU/mL and 5.7 Log10 U/mL of qHBsAg and qHBcrAg had a sensitivity of 71.4%, a specificity of 79.5%, a PPV of 65.2%, and an NPV of 83.8%, with an AUROC of 0.769; at 12 months, a combination of 3.8 Log10 IU/mL and HBcrAg 5.5 Log10 U/mL had a sensitivity of 73.7%, a specificity of 79.5%, a PPV of 63.6%, and an NPV of 86.1%, with an AUROC of 0.807. As the care of CHB shifts to medicines that give functional cures, they concluded that qHBsAg and qHBcrAg may be utilized to identify individuals who are unlikely to attain treatment endpoints.

Although the clearance rate is quite low, HBsAg clearance reflects a clinical cure. The objectives of the study by Cao *et al*[81] were the assessing the viability and safety characteristics of pegylated interferon α-2a (PEG-IFNα-2a) as a treatment alternative for dormant HBsAg carriers. A therapy group of 102 participants and a control group consisting of 42 subjects were formed from the 144 inactive HBsAg carriers who were

recruited. Subjects in the therapy group with HBV DNA < 20 IU/mL and 20 IU/mL ≤ HBV DNA < 2000 IU/mL, respectively, received PEG-IFNα-2a and PEG-IFNα-2a in combination with adefovir dipivoxil. There was a maximum of 96 wk in total for treatment. The therapy effectiveness was assessed using the seroconversion rates and HBsAg clearance at therapeutic weeks 48 and 96. According to protocol analysis, the therapy group's HBsAg clearance and seroconversion rates were 29.8% and 20.2% at week 48 and grew to 44.7% and 38.3% at week 96, respectively. Nonetheless, in weeks 48 and 96, the HBsAg clearance rate in the control group was 2.4%, and no patient attained seroconversion. HBsAg clearance was well predicted by the qHBsAg levels and variations at week 12 and week 24 of the therapy, as well as by the rise of ALT at week 12. The side effects matched those of therapy for long-term hepatitis B patients. They deduced that PEG-IFNα-2a-based therapies might attain greater amounts of HBsAg clearance, as measured by qHBsAg and seroconversion, and these treatments posed no risk to inactive HBsAg carriers.

**qAnti-HBc**

In HBV low-to-medium epidemic locations, anti-HBc has historically been used as a blood screening test and is thought to be an indication of prior or current HBV infections. Interestingly, most people with chronic or even cured HBV infection often have anti-HBc levels beyond the top limit of commonly used qualitative anti-HBc tests. In order to accurately establish the differences in anti-HBc levels across patients or the changes that occur with the course of the diseases, studies on the usefulness of anti-HBc levels in chronic HBV infection need repeated dilutions or assays with large dynamic ranges. Qualitative tests cannot determine these differences[82]. When it comes to CHB infections, those with active hepatitis in the immune-active phase usually have 10-fold greater levels of qAnti-HBc than people in the immune-tolerant or inactive-carrier phases when hepatitis is not present[83]. In individuals with CHB, there is a positive link

between serum ALT activity and qAnti-HBc, which suggests that the latter may be indicative of hepatic inflammation, according to many studies. Furthermore, it has been shown that qAnti-HBc favorably correlates with the liver biopsy-determined histological severity of hepatic inflammation. Surprisingly, research has shown that qAnti-HBc is linked to the severity of fibrosis and, when combined with other factors, may enhance the efficacy of non-invasive indices. However, the connection between fibrosis degree and qAnti-HBc is often smaller than the correlation with inflammation, indicating that the former relationship may be partly explained by the correlation between qAnti-HBc and liver inflammation[84-87]. The gold standard for identifying liver inflammation is a liver tissue biopsy, yet since it is invasive, it is not often performed. ALT, a frequently used marker for hepatocellular damage, has a risk of being ignored since moderate to severe hepatic inflammation is present in 20%-30% of individuals with normal ALT. Several studies have looked at the additional role of qAnti-HBc in identifying immune activation status and inflammation in HBV-positive persons with normal ALT levels[88,89]. Zhang *et al*[90] found that in 1376 untreated CHB patients with normal ALT, there was a dose-responsive association between the degree of liver inflammation and qAnti-HBc. The qAnti-HBc cut-off value for identifying moderate and severe inflammation was around 4.5 Log10 IU/mL. The investigation was conducted across many centers. This study also found a correlation between the reduction of liver histological inflammation and the drop in qAnti-HBc after NAs administration. Furthermore, Feng *et al*[91] similarly observed that substantial liver damage was associated with high qAnti-HBc in CHB patients with normal ALT; however, the study's cut-off value (3.7 Log10 IU/mL) seemed to be lower than the previous study. More specifically, a high level of qAnti-HBc was also observed to be related to considerable liver inflammation in populations of HBeAg-negative chronic HBV-infected individuals with normal ALT, which was evaluated by Yao *et al*[92].

The current qAnti-HBc assays are based on competitive/inhibitory, indirect, or double-antigen sandwich immunoassay technology and are available as enzyme-linked immunosorbent assays (ELISA), chemiluminescent microparticle immunoassays (CMIA), or CLEIA. Table 3 summarizes the techniques and units used to quantify anti-HBc. The development of the double-antigen sandwich immunoassay for qAnti-HBc was first established in 2010. This particular assay has been shown to exhibit enhanced sensitivity and specificity compared to other methods. However, it should be noted that the quantitation range of this test is very limited. The commercially available ELISA, which is based on the double-antigen sandwich immunoassay, may now be obtained from Wantai Biological Pharmacy Enterprise Company in Beijing, China. This ELISA can be calibrated against the WHO International Standard, allowing findings to be provided in IU/mL. The study indicated a linear detection range of 2–5 Log IU/mL, with a lower limit of quantitation of 0.1 IU/mL[93,94]. Lumipulse G Anti-HBc-N, a fully automated two-step sandwich CLEIA developed by Fujirebio in Tokyo, Japan, is another often-used test. The present methodology involves the automated reporting of anti-HBc IgG levels as the cut-off index (COI), which is determined by calculating a multiple of the cut-off value derived from calibration data. The lowest limit of quantification was declared by the manufacturer as 1 COI. However, it is possible to calibrate the data using the WHO International Standard in order to acquire IU/mL. The lower limit of detection was 0.5 IU/mL. In comparison to other approaches using chemiluminescence for detecting qAnti-HBc, this particular method exhibited superior levels of sensitivity, specificity, and a wider linear dynamic range[95]. Furthermore, for the purpose of determining the quantities of anti-HBc in serum, a straightforward and quick fluorescence point-of-care test based on a lateral flow immunoassay technique has been developed. The method used is a competitive time-resolved fluoroimmunoassay which utilizes microparticles of carboxylate-modified polystyrene Europium III chelate as a reporter. The fluorescence is measured using a portable TRF strip reader. The test

demonstrates a very low detection limit, as well as rapidity with a reported time of 15 min and cost-effectiveness[96].

## qAnti-HBc AND FUNCTIONAL CURE

In NA-suppressed CHB patients undergoing PEG-IFN-α add-on treatment, Wang *et al*[97] studied qAnti-HBc and qHBcrAg as predictors for HBsAg clearance. Following at least a year of NA therapy, 74 CHB patients with HBV DNA suppression (HBV DNA < 20 IU/mL) and quantitative HBsAg (qHBsAg) < 1500 IU/mL were included. While 59 individuals got PEG-IFN-α add-on treatment, 15 patients persisted on NA monotherapy. Every 12 or 24 wk, respectively, the add-on and NA-alone groups' serum qAnti-HBc and qHBcrAg levels were measured. They discovered a negative correlation between the length of previous NA treatment and baseline blood qAnti-HBc but not qHBcrAg levels. Both qAnti-HBc and qHBcrAg levels decreased after 48 wk of therapy, and in the add-on and NA groups, 17/59 (28.81%) and 0/15 (0%) of patients, respectively, achieved HBsAg clearance. Patients in the add-on group with baseline qAnti-HBc < 0.1 IU/mL had a substantially greater percentage of HBsAg clearance (52.63%). Baseline qAnti-HBc was shown to be an independent predictor of HBsAg loss using logistic regression analysis, but not qHBcrAg. The combination of qAnti-HBc and qHBsAg exhibited a greater predictive value for HBsAg clearance, according to an examination of the ROC curve. In NA-suppressed CHB patients undergoing PEG-IFN-α add-on treatment, they determined that a combination of qHBsAg and baseline qAnti-HBc levels would be a superior prediction technique for HBsAg clearance. During chronic HBV infection, the HBeAg-negative state is associated with a wide range of clinical conditions, from inactive carriers with overall survival comparable to HBV-non-infected individuals to active chronic hepatitis with significant hepatic necro-inflammatory activity and rapid progression to cirrhosis. The danger of transitioning from an inactive carrier status to an active infection is always unexpected. Current

antiviral therapy may change the course of an ongoing infection by halting fibrosis development and lowering the risk of end-stage consequences. Thus, distinguishing true inactive carriers from individuals with persistently low viremic active infection (HBV DNA > 2000 IU/mL with normal ALT) and predicting the potential of HBV DNA undetectability and HBsAg seroclearance are critical. Unlike in HBeAg-positive phases, a lower level of qAnti-HBc is linked with a better clinical result in HBeAg-negative phases[98]. Wu *et al*[99] conducted a study to investigate the potential of qAnti-HBc level as a biomarker for predicting the recurrence of CHB patients who had HBsAg clearance after antiviral therapy. Sixteen instances of recurrence and fifty-seven cases of non-recurrence out of the 73 patients with HBsAg clearance were included. Before and after treatment, the qAnti-HBc level was measured using a recently developed double-sandwich immunoassay. The predictive power of qAnti-HBc levels for recurrence was assessed using logistic regression analysis. They discovered that both the recurrence and non-recurrence groups' post-treatment qAnti-HBc levels were much lower than those from before therapy. Furthermore, compared to the non-recurrence group, the recurrence group's falling trend was much larger.

In 60 cases of CHB patients treated with PEG-IFN-α2a plus NAs antiviral therapy previously, Lin *et al*[100] investigated the potential role of qAnti-HBc in predicting HBsAg clearance (41 cases in the clearance group and 19 cases in the non-clearance group). The qAnti-HBc levels of the patients were measured using the double antigen sandwich technique at baseline, 24, 48, 72, and 96 wk. When antiviral treatment was prolonged, qAnti-HBc levels in both the clearance and non-clearance groups showed a progressive downward trend. However, at baseline and at subsequent detection time points during the antiviral treatment, the levels in the clearance group were significantly higher than those in the non-clearance group. At week 24, multivariate logistic regression revealed a reduction in both the baseline qAnti-HBc level and qHBsAg. The greatest independent predictor of HBsAg clearance among them was the

baseline qAnti-HBc level; its sensitivity and specificity for predicting HBsAg clearance over 3.40 Log10 IU/mL were 56.1% and 89.5%, respectively. To increase prediction accuracy, integrated predictors were constructed using the logistic regression model as a guide. The combined factor 3 had the best predictive value out of all of them. 80.5% and 78.9%, respectively, were the sensitivity and specificity. Furthermore, the HBsAg clearance predicted rate had reached 94.12%-100%, and the combined index — which is the combination of any two or more indices based on the baseline qAnti-HBc level — had further increased the predictive value. Lou *et al*[101] conducted a study to employ biomarkers to assess HBV reinfection in patients following orthotopic liver transplantation. They enlisted 79 individuals who had liver transplants, and biomarker levels were measured at various time periods. They discovered that 42 instances had HBsAg loss with a median time of 65.2 mo following liver transplantation, while 37 patients had HBsAg recurrence with a median time of 8.8 mo. The highest Youden's index values at the start of the study were 4.25 Log10 IU/mL for qAnti-HBc and 2.82 Log10 IU/mL for qHBsAg, with areas under the curves of 0.685 and 0.651, respectively. The Kaplan-Meier technique revealed that the levels of qHBsAg and qAnti-HBc were significantly different between the two groups. Furthermore, the Cox regression model verified the predictive efficacy of qAnti-HBc at baseline. They hypothesized that lower pre-transplantation qAnti-HBc levels are linked to HBV re-infection. The baseline concentration of qAnti-HBc in patients obtaining liver transplantation is a potential predictor of HBV recurrence and may be used to guide antiviral therapy for HBV infection.

In individuals receiving antiviral therapy, baseline anti-HBc levels may be predictive of HBeAg seroconversion. A greater baseline concentration of qAnti-HBc was a robust predictor of HBeAg seroconversion in a retrospective investigation of 231 patients treated with pegylated interferon and 560 receiving NAs, outperforming both HBV DNA and ALT levels. Patients receiving NAs therapy had a much higher reduction in

anti-HBc levels following treatment than those getting pegylated interferon, which might be attributed to interferons' stronger pleiotropic effects on immune activation[102]. QAnti-HBc levels at the end of treatment may also be indicative of viral recurrence after stopping NAs therapy. Indeed, after controlling for age, HBeAg status, and length of consolidation therapy, high levels of qAnti-HBc at the end of treatment were linked with a lower probability of clinical relapse. Further stratification of anti-HBc levels at the completion of therapy revealed that levels of 1000 IU/mL were linked with the lowest likelihood of clinical relapse (21% at year 4), compared to values between 100 and 999 IU/mL (50% at year 4) and 100 IU/mL (85% at year 4)[103].

## CONCLUSION

Anti-HBc IgM and IgG antibodies are the most common serological markers of HBV infection. Anti-HBc IgM is positive during the inflammatory phase of the liver but turns negative during the recovery stage; hence, it may assist in identifying acute HBV infection or acute exacerbation from quiescent CHB. Anti-HBc IgG is a marker for present and prior HBV infection that may last a lifetime. The double-antigen sandwich approach was utilized to quantify anti-HBc antibodies, and the detection indicated the sum of IgG and IgM antibodies. HBsAg reveals the transcriptional activity of both cccDNA and integrated HBV DNA, and the continued seroclearance of high-sensitivity or ultra-sensitive qHBsAg may signify a complete cure of CHB to some extent. HBV antigens, particularly HBsAg, are also implicated in the immunopathogenesis of hepatitis B. Thus, HBsAg loss may dramatically improve aberrant immune function, which may simplify the clearance of remaining viruses such as cccDNA and integrated HBV DNA. Some outstanding concerns must be resolved. It is uncertain if continuous HBsAg loss indicates that cccDNA and integrated HBV DNA are fully inactive, preventing HBsAg expression, or that the majority of them are removed. Moreover, a growing number of novel medications that suppress HBV and improve host immune

responses have been established in numerous clinical studies. Beyond the prognostic and diagnostic utility of qAnti-HBc levels, the actual process remains unknown. The level of qAnti-HBc is recognized as a surrogate measure of HBV-specific adaptive immune response activity. However, the increased immune response is a double-edged sword since it produces severe liver damage in an effort to eliminate the infection. HBV core antibody quantification is a novel, non-invasive biomarker that may be utilized to address a variety of diagnostic challenges. It may offer useful information on viral activity, disease advancement, and therapy responses. qAnti-HBc, like other recently used biomarkers, cannot be relied on as a single diagnostic test to address all problems. Based on existing evidence, it is clear that when paired with qHBsAg, its diagnostic and prognostic efficacy may be considerably increased. More studies with larger and more diverse patient groups are needed to evaluate the true utility of these biomarkers.

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