

Update on HIV/HBV Coinfection: Pathogenesis and Treatment

Marion Peters, MD

Professor of Medicine and Chief of Hepatology Research
University of California, San Francisco, San Francisco, California

SUMMARY BY TIM HORN

EDITED BY JAMES F. BRAUN, DO; STEPHEN LOCARNINI, BSc(HONS), MBBS, PhD, FRC(PATH);

JOE SASADEUSZ, MBBS(HONS), PhD, FRACP, FRCC

Reprinted from *The PRN Notebook*, DECEMBER 2002.

Dr. James F. Braun, Editor-in-Chief. Tim Horn, Executive Editor.

Published in New York City by the Physicians' Research Network, Inc.,

John Graham Brown, Executive Director. For further information and other articles available online, visit <http://www.prn.org> All rights reserved. ©DECEMBER 2002.

IT HAS BEEN ESTIMATED THAT APPROXIMATELY 250 million people worldwide have chronic hepatitis B virus (HBV) infection. In the United States alone, an estimated 1.25 million people—0.35% of the U.S. population—have chronic HBV, defined as patients with a positive hepatitis B surface antigen (HBsAg) serology for more than six months. While hepatitis B vaccination programs are an important component of hepatitis B prevention strategies, they will not have an impact on those already living with this potentially fatal disease.

Carriers of HBV are at increased risk of developing cirrhosis, hepatic decompensation, and hepatocellular carcinoma. Although most carriers do not develop hepatic complications, 25% to 40% do go on to develop serious HBV-related manifestations during their lifetime.

While chronic HBV disease in the setting of HIV has not yet been listed as an AIDS-defining illness by the U.S. Centers for Disease Control, it is undoubtedly an opportunistic infection and not uncommon in HIV-positive individuals because of similar modes of transmission. Between 90% and 95% of HIV-positive people have serologic evidence of past HBV infection, and approximately 15% are chronic carriers.

HIV-positive individuals, particularly those with suppressed immune systems, are less likely to respond to vaccination against hepatitis B and are more likely to develop chronic disease after being exposed to the virus. In addition, individuals coinfecting with HIV and HBV are more likely to present with atypical serologies, to have higher HBV-DNA levels, and to experience more profound liver disease as a result of chronic infection.

This article reviews much of the current thinking surrounding the pathogenesis, diagnosis, monitoring, and treatment of chronic hepatitis B in HBV-monoinfected and HIV/HBV-coinfecting patients. What follows is an expanded summary of a provocative and eloquent PRN lecture delivered by Dr. Marion Peters, Chief of Hepatology Research at the University of California, San Francisco.

Virology of HBV

HBV BELONGS TO A FAMILY OF DNA VIRUSES called hepadnaviruses. When examined under an electron microscope, intact HBV virions—the Dane particles—appear as spheres 42 nm in diameter (see Figure 1). Each complete virus consists of an inner core (nucleocapsid or hepatitis B core antigen [HBcAg]) surrounded by an outer protein coat or envelope, the hepatitis B surface antigen [HBsAg]. HBsAg is a 22 nm tubule—or sphere when visualized end on—and is produced in great excess to Dane particles.

The HBV genome is a circular, partially double-stranded DNA of approximately 3,200 base pairs. There are four overlapping reading frames, which encode the envelope, precore/core, polymerase, and X proteins. The envelope open reading frame encodes the large, middle, and small surface glycoproteins of HBsAg. The precore/core open reading frame is translated into a precore polypeptide, which is secreted as hepatitis B “e” antigen (HBeAg) and into HBcAg. It is important to note here that mutations in the core promoter and precore region can result in decreased production or loss of serum HBeAg with continued production of core antigen,

which is incorporated into the HBV virion. The significance of these mutations is discussed in greater detail below.

Upon entering hepatocytes, the HBV genome is transported to the nucleus and is converted to covalently closed circular DNA (cccDNA). The HBV cccDNA serves as the template for transcription of mRNA and the RNA pregenome. Once transferred to the cytoplasm, HBV polymerase uses reverse transcription to convert the RNA pregenome into new circular genomic DNA.

HBV is not a cytopathic virus. Replicative HBV, either during the acute or chronic stages of infection, causes liver disease because of vigorous cytotoxic T-lymphocyte (CTL) and cytokine responses directed at HBV-infected hepatocytes.

Natural History of HBV: Acute Infection

HBV INFECTION BEGINS WITH AN ACUTE PHASE, which can manifest itself as asymptomatic infection, acute hepatitis, or rarely fulminant liver failure. Asymptomatic infection, the most common acute infection course, begins with active HBV replication in hepatocytes. Detectable HBV-DNA levels in peripheral blood are initially low and limited in time and may precede mild or moderate elevations in serum transaminases. HBsAg and HBeAg become detectable in blood samples—the core antigen cannot be detected in peripheral blood samples, only in hepatocytes—within two to eight weeks and can persist for several weeks. IgM antibodies to HBcAg occur early and decrease after six months, but total anti-HBc remains for life. Anti-HBs becomes positive after loss of active viral replication, usually after six months (see Figure 2).

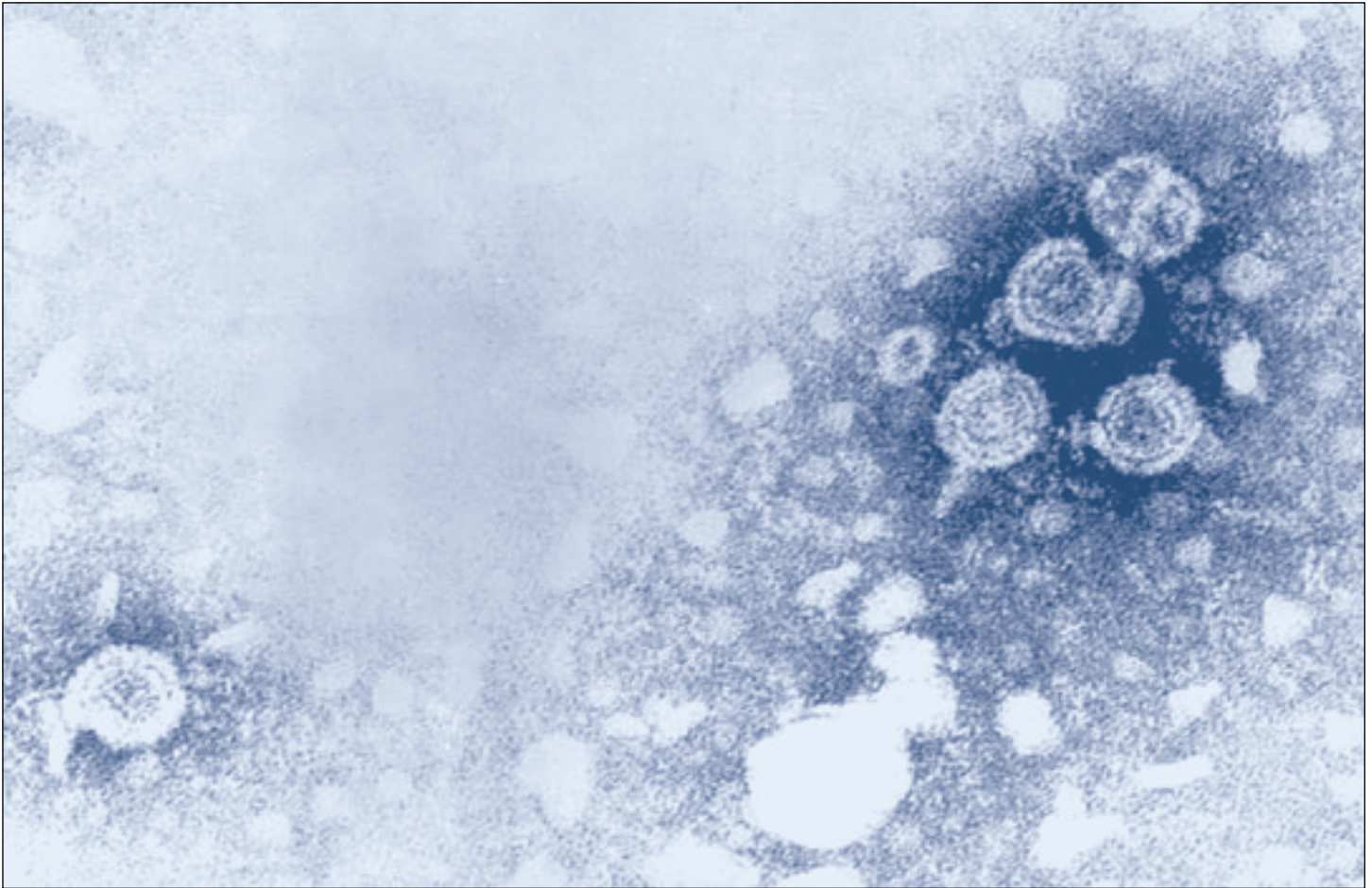


FIGURE 1. Electron Micrograph of Hepatitis B Virions

Transmission electron micrograph of hepatitis B virions. Intact HBV virions—the Dane particles—appear as spheres 42 nm in diameter. Each complete virus consists of an inner core (nucleocapsid or hepatitis B core antigen [HBcAg]) surrounded by an outer protein coat or envelope, the hepatitis B surface antigen [HBsAg]. HBsAg is a 22 nm tubule—or sphere when visualized end on—and is produced in great excess to Dane particles.

Source: Public Health Image Library, U.S. Centers for Disease Control. Photograph by Erskine Palmer, MD.

Clinical disease in acute HBV infection may be asymptomatic, mild, moderate or severe. Symptomatic HBV infection can exist in the icteric and anicteric forms. Anicteric infection is typically associated with a flu-like illness without jaundice. Icteric infection is associated with symptoms ranging from mild jaundice to nonfatal subacute hepatic necrosis (3% of all acutely infected patients), to fatal fulminant hepatitis (less than 1% of all acutely infected patients). In those with acute hepatitis, HBV-DNA levels, along with serum transaminases, may be high and prolonged. Serum levels of HBeAg and HBsAg may remain detectable for several months before antibodies replace them.

Regardless of the clinical course taken, most HBV-infected adults are capable of generating broad-based CTL and antibody responses to the virus. Once these develop,

the infection becomes latent, with serum evidence of immunity, which usually remains with the patient for the rest of his or her life. Under unusual circumstances, such as immune suppression or organ transplantation, HBV can be reactivated, as intact virions may be hidden from the immune system in hepatocytes.

Less than 5% of adult HBV-infected people develop chronic disease—that is, remain HBsAg-positive longer than six months after infection. However, as pointed out in practice guidelines published by the American Association for the Study of Liver Disease (AASLD) in the December 2001 issue of *Hepatology*, it may take a few more months for some individuals to clear HBsAg, but HBsAg should be undetectable one year after acute HBV infection (Lok, 2001).

The outcome of acute HBV infection is

largely determined by host factors, particularly age and immune competence at the time of exposure. In infants exposed to HBV at birth or shortly thereafter, chronic infection is established over 90% of the time. The risk of chronic HBV infection declines as the exposed individual increases in age, falling to 25% to 50% in young children, to 1% to 5% of people exposed during adulthood. In adults, the risk of chronic infection is largely dependent on the general health of the immune system; hence the increased prevalence of chronic infection among transplant patients, patients receiving hemodialysis, patients receiving corticosteroid treatment, and HIV-infected patients.

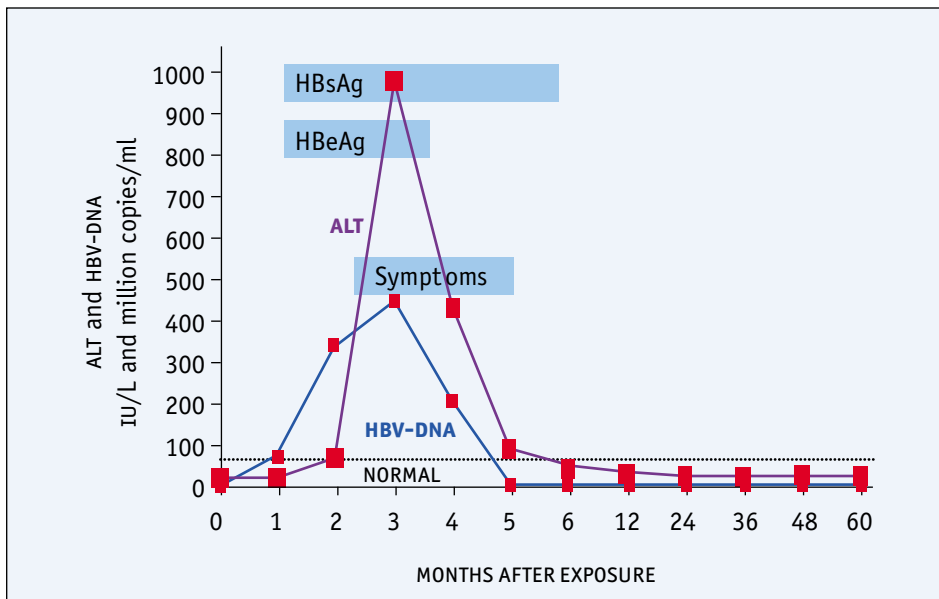


FIGURE 2. Typical Acute Hepatitis B

HBV infection begins with an acute phase, which can manifest itself as asymptomatic infection, acute hepatitis, or rarely fulminant liver failure. Asymptomatic infection, the most common acute infection course, begins with active HBV replication in hepatocytes. Detectable HBV-DNA levels in peripheral blood are initially low and limited in time and may precede mild or moderate elevations in serum transaminases. HBsAg and HBeAg become detectable in blood samples—the core antigen cannot be detected in peripheral blood samples, only in hepatocytes—within two to eight weeks and can persist for several weeks. IgM antibodies to HBcAg occur early and decrease after six months, but total anti-HBc remains for life. Anti-HBs becomes positive after loss of active viral replication, usually after six months.

Source: Marion Peters, MD

Natural History of HBV: Chronic Infection

THE NATURAL HISTORY OF CHRONIC HBV CAN be broken down into three distinct phases: immune tolerance, immune clearance, and latency (nonreplicative). In general, those infected with HBV at birth are the only patients to enter the immune-tolerance phase, characterized by the presence of HBeAg, high HBV-DNA titers, and normal ALT levels. Adults who remain chronically infected after acute exposure typically shift immediately into the immune-clearance phase (infants with chronic infection enter the immune-clearance phase later on in life). During the immune-clearance phase of chronic HBV infection, HBeAg is usually present, HBV-DNA levels are elevated (above 100,000 copies/mL), and ALT/AST levels may be either persistently or intermittently elevated (see Figure 3).

Hepatic flares—characterized by spikes in HBV-DNA and ALT levels—are common during the immune-clearance phase. These spikes indicate that the immune

system is making regular attempts to eliminate HBV from the infected hepatocytes. With each flare, HBV-DNA is decreased and there is, over time, a spontaneous HBeAg to anti-HBe shift. Once this occurs, chronically infected patients move into the latent phase—or the inactive carrier state—of infection, in which transaminases stabilize and symptoms are minimal.

It is vital to point out that the absence of HBeAg is not necessarily synonymous with inactive infection. HBeAg-negative chronic HBV infection, characterized by ongoing HBV-DNA replication and continued necroinflammation in the liver, has been reported in all parts of the world and is rapidly becoming the most prevalent form of liver disease in some parts of the world (Dr. Stephen Locarnini, personal communication). The vast majority of HBeAg-negative chronic hepatitis B harbor HBV variants in the precore open reading frame or basic core promoter (BCP) region, the most common being G1896A in the precore region. This mutation creates a premature stop

codon in the precore region, which hinders production of HBeAg. It is most commonly associated with HBV genotype D, which is prevalent in the Mediterranean basin and parts of Asia, and is rarely detected in patients with HBV genotype A, which is the most prevalent strain in the United States. Patients with HBeAg-negative chronic HBV infection tend to have lower HBV-DNA levels and are more likely to run a fluctuating course characterized by persistently elevated or fluctuating ALT levels.

Overall, the clinical course of chronic infection is variable and ranges from mild to severe. On average, patients with chronic HBV infection maintain high HBV-DNA levels for five to ten years and a detectable HBV-DNA viral load for 20 years. Approximately 0.5% to 1% of HBsAg carriers clear HBsAg yearly, most of whom will seroconvert to anti-HBs. There is a 25% to 40% lifetime risk of end-stage liver disease in chronically infected patients. Hepatocellular carcinoma (HCC) is statistically more likely to occur in chronically infected men and develops in approximately 2-4% of patients per year. And because HBV has been found to be an oncogenic virus, HCC can occur in the absence of cirrhosis.

The Significance of Coinfection

AS EXPLAINED ABOVE, ONLY A SMALL PERCENTAGE of adult-acquired HBV develops into chronic infection. This, however, is not the case in HIV-infected adults: Between 10% and 15% of patients coinfecting with both viruses develop chronic hepatitis B. With respect to prevalence rates, a study presented at the 9th Conference on Retroviruses and Opportunistic Infections provided a glimpse at the magnitude of the coinfection problem in the United States (Thio, 2002). A large cohort of 5293 gay men was followed prospectively, and mortality was assessed based on HBV serologic status. A total of 326 (6%) men were HBsAg positive and, of these, 213 (65%) were HIV-positive. Of the 4967 HBsAg-negative men, 2346 (47%) were HIV-positive. The liver-related mortality rate was 1.1/1000 person-years, and was higher in the HBsAg-positive than HBsAg-negative individuals. When liver-attributable mortality was examined in those who were HIV-infected, coinfection with HBV significantly increased the risk (14.2/1000 person-years) compared with those with HIV alone (1.7/1000 person-years) or HBV

alone (0.8/1000 person-years). hcv infection was found in only 7% of HIV/HBV coinfecting patients who experienced liver-related deaths. Prior to 1996—the introduction of highly active antiretroviral therapy (HAART)—the liver-related mortality rate among HIV-positive patients was 2.5/1000 person-years. After 1996, the liver-related mortality rate increased to 4.0/1000 person-years. This is in sharp contrast to the AIDS-related mortality rate, which strongly decreased after 1996.

Patients coinfecting with HIV and HBV often present with atypical serological test results. This was the experience of investigators associated with the Swiss HIV Cohort Study, in an analysis involving frozen serum specimens that were sequentially obtained over time from a cohort of 57 HIV-infected patients (Hofer, 1998). All tested positive only for anti-HBc, and were retested for other HBV markers, including HBV-DNA (Hofer, 1998). During a median of 31 months from the first to the last serum collected, anti-HBc remained the sole marker of HBV infection in 98.2% of the patients. PCR to detect HBV for HBV core and HBV surface gene was positive in 126 (62.4%) and 121 (59.9%) of all 202 serum samples, respectively. Over time, HBV-DNA was detected at least once in 51 (89.5%) patients. In contrast, decomplexed HBsAg was detected at least once in 14 (24.6%) patients. Among patients positive for HBV-DNA and negative for HCV antibodies, 8/22 (36.4%) had necroinflammatory disease that was attributable only to persisting HBV infection. Similarly, 12/29 (41.4%) patients positive for both HBV-DNA and HCV had chronic viral hepatitis, but their ALT values were significantly higher. In other words, in HIV-infected patients, the detection of anti-HBc—as opposed to HBsAg—may be indicative of chronic HBV infection, and is in part associated with chronic hepatitis and ALT elevation. “It is important that HBV status be obtained at baseline for all HIV-positive patients,” commented Dr. Peters. “This should include HBsAg, anti-HBs, and total anti-HBc serologies. Patients who test HBsAg-positive should then have HBeAg, anti-HBe, and HBV-DNA tests performed to determine the state of infection and the need for antiviral therapy for HBV.”

It is not entirely understood what impact HIV has on the severity of chronic HBV infection. Dr. Peters commented that, as HIV disease progresses, cellular and humoral

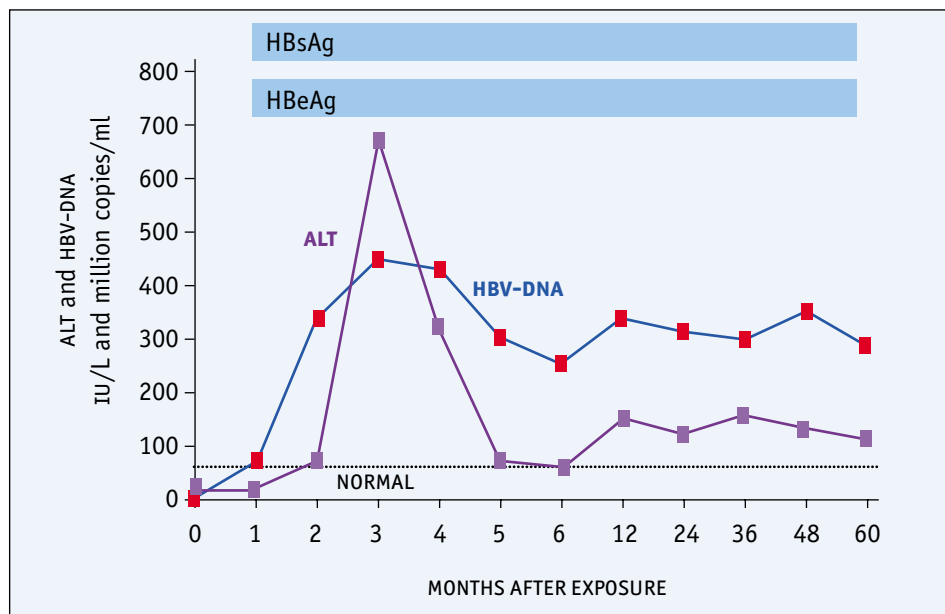


FIGURE 3. Typical Chronic Hepatitis B

The natural history of chronic HBV can be broken down into three distinct phases: immune tolerance, immune clearance, and latency (nonreplicative). In general, those infected with HBV at birth are the only patients to enter the immune-tolerance phase, characterized by the presence of HBeAg, high HBV-DNA titers, and normal ALT levels. Adults who remain chronically infected after acute exposure typically shift immediately into the immune-clearance phase (infants with chronic infection enter the immune-clearance phase later on in life). During the immune-clearance phase of chronic HBV infection, HBeAg is usually present, HBV-DNA levels are elevated (above 100,000 copies/mL), and ALT/AST levels may be either persistently or intermittently elevated.

Source: Marion Peters, MD

immunity to HBV gradually decreases or is sometimes lost. “There is a balance between the immune response and liver damage: where there is no immune response to HBV, there is no ongoing liver damage,” she said. “But HBV replication may be enhanced. Then again, where there is an active immune response, there will be ongoing liver damage. Thus, patients who have not yet started HAART may have high HBV-DNA but no liver damage. Once they are treated with HAART and develop healthy CD4+ cell counts, they may develop severe liver damage and even liver failure.”

In general, patients with HIV/HBV coinfection tend to have higher levels of HBV-DNA, lower rates of spontaneous HBeAg seroconversion, and more severe liver disease. Dr. Peters also pointed out that signs and symptoms of liver disease in HIV/HBV coinfection can be somewhat confusing in the setting of HAART. First, the ability of HAART to spark the immune system—particularly if effective anti-HBV therapies are not included in the regimen—can result in a reactivation of necroinflammatory dis-

ease. There is also the issue of hepatotoxicity to consider, especially with the non-nucleoside reverse transcriptase inhibitors and protease inhibitors.

Treating Chronic HBV

THE PRIMARY GOAL OF TREATING CHRONIC HBV infection is to halt progression of liver disease by suppressing viral replication. Even though HBV is not a cytopathic virus, the immune system continues to respond and cause damage if the virus continues to replicate. In essence, persistent viremia portends liver disease and, as is the case with HIV, therapy should focus on halting viral replication.

As highlighted in the AASLD practice guidelines (Lok, 2001), the endpoints used to assess treatment response include normalization of serum ALT levels, undetectable HBV-DNA in peripheral blood using an unamplified assay, loss of HBeAg with or without detection of anti-HBe, and improvements in liver histology. The practice guidelines also point out that it is difficult

to compare response rates in clinical trials of the treatment of chronic HBV infection, as there have long been inconsistencies in the definition of response, lack of standardization of HBV viral load assays, and significant baseline differences in the patient populations studied. Still, improvements have been made in the therapy of chronic HBV infection, most notably the addition of adefovir dipivoxil to the therapeutic armamentarium.

Dr. Peters was quick to point out that the success of therapy is highly dependent on baseline transaminases, even when using nucleoside/nucleotide analogues. "High ALT levels indicate that an immune response is occurring in the liver. Antiviral agents will decrease the level of HBV but the immune response is required to shut off viral replication." Generally speaking, anti-HBV therapies are most efficacious in patients with ALT levels greater than two times the upper normal limit.

This is important to keep in mind when treating HIV and HBV-coinfected patients. Patients with lower CD4+ cell counts tend to have lower ALT levels—which can be tied to decreased coordination of CTLs associated with dwindling CD4+ cell activity. This speaks to the importance of implementing anti-HIV/anti-HBV drug regimen at the same time to decrease HBV viral load and to optimize the ability to induce an HBV CTL response when HIV is more controlled.

Interferon-Alpha

INTERFERON- α HAS BEEN APPROVED FOR MORE than a decade. Used at a dose of 5 million units every day or 10 million units three times weekly for 16 weeks, interferon- α monotherapy is associated with HBeAg clearance in approximately 30% of immunocompetent patients and HBsAg clearance in up to 10% of patients. Studies have also suggested that it is effective in reducing HBV-DNA levels and normalizing ALT levels in patients with HBeAg-negative chronic hepatitis B, although high rates of relapse are frequently seen. Interferon- α is less effective in patients with high baseline HBV-DNA levels and patients with normal ALT levels prior to initiating treatment, which is often seen in HIV/HBV coinfected individuals.

Unfortunately, the drug is associated with significant side effects including fever, myalgia, thyroid abnormalities, bone marrow suppression, and a litany of psychiatric symptoms. What's more, the drug

must be used cautiously in patients with cirrhosis, as it may exacerbate the immune response to the virus and lead to decompensation. Patients most likely to benefit from interferon- α therapy are those with normal synthetic function, no cytopenias, elevated ALT, consistent liver biopsy results, and no decompensation.

Dr. Peters spent some time reviewing the results of a study designed to better define the long-term prognosis of interferon- α treatment (Lau, 1997). The study followed 103 patients with chronic HBV infection who underwent interferon- α therapy in three clinical trials between 1984 and 1991. Follow-up parameters included serological status, biochemical evidence of liver disease, and liver complications or mortality through 1994. Consistent with the results of several interferon- α clinical trials, 31/103 (30%) responded to therapy with loss of HBeAg and HBV-DNA. Responders were more likely than nonresponders to be women, black, and to have more severe liver disease including cirrhosis. Up to 11 years—with a mean of 6.2 years—after therapy, a higher percentage of responders than nonresponders were still negative for HBeAg (94% vs. 40%) and HBsAg (71% vs. 8.3%). Overall, the rate of liver-related complications and death did not differ by interferon- α response, but with statistical adjustments made for cirrhosis, nonresponders had higher rates of liver-related complications and mortality. In other words, the response to interferon- α therapy in chronic hepatitis B infection is usually a sustained improvement in disease markers and, when cirrhosis is considered, patient outcome.

In HIV/HBV-coinfected patients, only those with CD4+ counts greater than 350 to 400 CD4+ cells stand a good chance at an effective response (Wong, 1995; MacDonald, 1987).

Pegylated Interferon

BECAUSE OF THE IMPROVED RESULTS OF pegylated versions of interferon- α over standard versions of interferon in the management of chronic hepatitis C infection, both Schering-Plough and Roche Pharmaceuticals—the manufacturers of Peg-Intron- and Pegasys-brand pegylated interferons, respectively—have their eyes set on an expanded indication to include patients with chronic HBV infection.

As described by Dr. Peters, a clinical tri-

al of Pegasys was carried out by Dr. Graham Cooksley of the Royal Brisbane Hospital Research Foundation in Brisbane, Australia, and his colleagues and reported this past spring at the 37th Annual Meeting of the European Association for the Study of the Liver (EASL) in Madrid (Cooksley, 2002). This was a four-arm study using three different doses of Pegasys—90 μ g, 180 μ g, and 270 μ g, weekly—compared with standard interferon over a period. The primary endpoint of the study was loss of HBeAg, a decrease in HBV-DNA to levels below 500,000 copies/mL, and normalization of ALT after six months of treatment.

A manuscript is currently in preparation, but the results indicated that more than twice as many patients (28%) receiving Pegasys 180 μ g weekly for six months responded favorably, compared with patients (12%) receiving standard interferon. As a result of these encouraging results, two further phase III trials are under way using Pegasys 180 μ g weekly in HBeAg-positive and HBeAg-negative chronic hepatitis B. Both of these studies compare pegylated interferon to lamivudine and to a combination of both lamivudine and pegylated interferon.

Lamivudine

GLAXOSMITHKLINE'S LAMIVUDINE, FIRST approved for the treatment of HIV in the mid-1990s, was awarded a second FDA approval for the treatment of chronic HBV infection in December 1998 (EpiVir-HBV). The dose of lamivudine typically used to treat chronic HBV is 100 mg once daily, compared to the 150 mg twice daily and 300 mg once daily schedules used to treat HIV. If used to treat HIV/HBV-coinfected patients, the 150 mg BID/300 mg QD dosing schedule should be used.

In patients with HBeAg-positive chronic HBV infection who have persistent or intermittent ALT elevations, three clinical trials involving a total of 731 treatment-naive patients who received lamivudine for one year reported that HBeAg seroconversion (including loss of HBV-DNA based on non-PCR assay) occurred in 16% to 18% of patients compared with 4% to 6% of untreated controls ((Dienstag, 1999; Lai, 1998; Schalm, 2000). Histologic improvement, defined as a reduction in necroinflammatory score greater than two points, was observed in 49% to 56% of treated patients and in 23% to 25% of controls.

In patients with HBeAg-negative chronic HBV infection who have elevated ALT levels prior to starting therapy, one study demonstrated a virologic and biochemical response in 34/54 (63%) patients who received 24 weeks of lamivudine therapy, compared to 3/53 (6%) of patients who received placebo (Tassopoulos, 1999). Of the 54 patients who completed one year of lamivudine treatment, HBV-DNA was undetectable by bDNA assay in 65% and by PCR assay in 39% of patients, and histologic improvement was seen in 60% of patients. Other studies have reported similar findings, although it should be noted that the vast majority of patients relapsed when lamivudine treatment was stopped.

Dr. Peters pointed out that HBeAg seroconversion, with reductions in HBV-DNA, is more likely to occur over time with lamivudine therapy. A study in Chinese patients with chronic hepatitis B showed that treatment with lamivudine for one year significantly improved liver histology and enhanced HBeAg seroconversion compared with placebo (Leung, 2001). Fifty-eight patients from this one-year study have received long-term treatment with lamivudine 100 mg. After three years of continuous treatment with lamivudine 100 mg daily, 23/58 (40%) patients achieved HBeAg seroconversion. In patients with baseline serum ALT greater than two times the upper normal limit, the rate of HBeAg seroconversion was 65%. Median HBV-DNA concentrations were below the level of detection, and median ALT concentrations were within the normal range throughout the three years of treatment.

While long-term therapy with lamivudine increases the possibility of HBeAg seroconversion, it also comes with the risk of developing key mutations in the YMDD motif, a highly conserved domain of all HBV reverse transcriptases required for HBV-DNA polymerase activation. The primary mutation associated with drug resistance is M552I/V—the equivalent of the M184V mutation associated with HIV resistance to lamivudine.

The clinical significance of lamivudine resistance in chronic HBV infection is not well understood. Follow-up data from clinical trials have demonstrated that resistance mutations occurred in 15% to 32% of patients treated with lamivudine for 52 weeks and as many as 67% of HBV-monoinfected patients (Lai, 2000) and 90% of HIV/HBV-coinfected patients (Benhamou,

1999) treated with lamivudine for a total of four years. Yet, some patients with lamivudine resistance mutations continued to experience HBeAg seroconversions, partial suppression of HBV-DNA, and improved biochemical and histologic parameters, likely because of decreased replicative capacity associated with HBV resistance to lamivudine (Leung, 2001). On the flipside, some strains of lamivudine-resistant HBV are fully replication competent and pathogenic. There can also be exacerbations of underlying liver disease in patients with HBV-lamivudine resistance. Moreover, HBeAg seroconversions are not always durable, and there is still a significant amount of HBV replication going on in the setting of lamivudine resistance.

Adefovir Dipivoxil

THE MOST RECENT ADDITION TO THE HBV therapeutic landscape is adefovir dipivoxil (Hepsera). It is being manufactured by Gilead Sciences and is the same drug that was studied previously, at higher doses, as a treatment for HIV, and subsequently rejected by the FDA because of high rates of renal toxicity—proximal renal tubular necrosis—at the 60 mg dose studied. The FDA-approved 10 mg adefovir dipivoxil dose has not been associated with renal toxicity, at least not in patients with normal renal function prior to initiating therapy.

Laboratory evaluations have suggested that the drug is effective against both wild-type and lamivudine-resistant HBV. There have also been a handful of phase I and phase II studies demonstrating that adefovir dipivoxil is associated with a 4 log reduction in HBV-DNA and is associated with HBeAg to anti-HBe seroconversion rates of 20% to 27% (Gilson, 1996; Heathcote, 1998; Jeffers 1998). It was the successful completion of two phase III studies involving patients with both HBeAg-positive and HBeAg-negative chronic hepatitis B that led to the approval of adefovir dipivoxil earlier this year. Highlights from these two studies, as well as those from a small open-label study involving HIV/HBV-coinfected patients, are reported here.

Study 437 was a randomized, double-blind, placebo-controlled phase III clinical trial evaluating the safety and efficacy of adefovir dipivoxil once daily as monotherapy compared to placebo in patients with HBeAg-positive chronic HBV infection. The study enrolled 515 patients in the United

States, Canada, Europe, Australia and Southeast Asia. Forty-eight-week results were first presented at the 52nd Annual Meeting of the American Association for the Study of Liver Diseases (AASLD), held in June 2001, in Dallas (Marcellin, 2001).

In this study, two doses of adefovir dipivoxil were evaluated: the 10 mg dose for which Gilead currently has approval and an exploratory 30 mg dose. During the first year of Study 437, 172 patients were randomized to receive adefovir dipivoxil 10 mg, 173 to receive adefovir dipivoxil 30 mg and 170 to receive placebo.

For patients with assessable baseline biopsies, improvement in liver histology was observed in 53% of patients treated with 10 mg adefovir dipivoxil, compared to 25% of the placebo-treated patients. In addition to improvement in liver histology, HBeAg-to-anti-HBe seroconversion was observed in 12% of patients treated with 10 mg adefovir dipivoxil for 48 weeks, compared to 6% of patients on placebo.

Reductions in HBV-DNA also were observed. Following 48 weeks of treatment, patients in the 10 mg adefovir dipivoxil group had a median reduction in HBV-DNA from baseline of 3.52 log, compared to a reduction of 0.55 log in patients receiving placebo. Additionally, a median reduction in ALT levels of 51 IU/L was observed in the 10 mg adefovir dipivoxil group, compared to a reduction of 17 IU/L in the placebo group. Forty-eight percent of patients treated with adefovir dipivoxil achieved normalization of ALT levels, compared to 16% of patients receiving placebo.

Gilead Study 438 was an international, multicenter, double-blind, placebo-controlled phase III clinical trial that enrolled 185 patients with HBeAg-negative chronic HBV infection and compensated liver function. The study was conducted in Australia, Canada, France, Greece, Israel, Italy and Southeast Asia. Patients were randomized to receive adefovir dipivoxil 10 mg once daily or placebo for 48 weeks. Data were presented at the 37th EASL in Madrid (Hadziyannis, 2002).

Liver biopsies were obtained from 178 patients. Sixty-four percent of patients treated with adefovir dipivoxil exhibited significant improvement in liver histology, compared with 33% of patients who received placebo. Improvement was defined as a greater than or equal to two-point reduction in the Knodell HAI score—a mea-

sure of necroinflammation—with no concurrent worsening of fibrosis. In terms of virologic improvements, 48 weeks of adefovir dipivoxil treatment was associated with a reduction in serum HBV-DNA from baseline of 3.91 log, compared with a median reduction of 1.35 log among patients who received placebo. More importantly, HBV-DNA were undetectable (<400 copies/mL) in 51% of patients who received adefovir dipivoxil, whereas none of the patients in the placebo group had undetectable HBV viral loads after 48 weeks. Improvements in biochemical parameters were also reported: ALT levels normalized in 72% of the patients receiving adefovir dipivoxil, compared with 29% of the patients who received the placebo.

The most common adverse events reported in both were headache, pharyngitis, and abdominal pain, although incidence rates of adverse events—including grade 3 and 4 laboratory abnormalities—were similar in both groups.

The resistance profile of adefovir dipivoxil was characterized in a poster presentation at the 37th EASL (Yang, 2002). The week-48 resistance surveillance included 695 chronic hepatitis B patients—all of whom participated in either Study 437 or Study 438—467 of whom received adefovir dipivoxil and 228 of whom received placebo. Due to undetectable serum HBV-DNA by PCR at 48 weeks, 197 adefovir dipivoxil-treated patients were not genotyped. Adefovir-associated resistance mutations were not observed in any of the patients evaluated.

The prolonged resistance profile and antiviral response of adefovir dipivoxil were further characterized at the 37th EASL in a poster presentation by Dr. Jenny Heathcote of Toronto Western Hospital and her colleagues (Heathcote, 2002). The presentation highlighted long-term resistance, efficacy, and safety data from the extension phase of Gilead's Study 412. In this phase II study, 39 patients with chronic hepatitis B who had previously been treated with adefovir dipivoxil 30 mg for >40 weeks received adefovir dipivoxil 10 mg. Of these patients, 11 had HBeAg-negative chronic hepatitis.

In patients treated with adefovir dipivoxil 10 mg beyond 48 weeks, adefovir dipivoxil was associated with a significant median reduction in serum HBV-DNA of 3.40 log that remained durable up to nearly two years (3.36 log at 100 weeks). Furthermore,

by week 100, HBV-DNA was undetectable (<400 copies/mL) in 70% of patients. Median reductions in ALT levels improved from week 48 to week 100 (36 IU/L to 48 IU/L), at which time these levels normalized in 63% of patients. In addition, over the course of the study, 21% of patients achieved HBeAg seroconversion. Finally, genotypic and phenotypic analyses revealed no adefovir-associated resistance mutations in patients who received 72 to 136 weeks of treatment with adefovir dipivoxil—encouraging results, indeed, but as in the HIV paradigm, resistance is likely inevitable.

Last but not least, it is important to mention a small open-label study of adefovir dipivoxil conducted in HIV/HBV-coinfected patients with lamivudine-resistant HBV (Benhamou, 2001). This study enrolled 35 coinfecting patients receiving lamivudine therapy (150 mg BID) as part of their anti-HIV regimen. Patients received a 10 mg once-daily dose of adefovir dipivoxil for 48 weeks while maintaining their existing antiretroviral regimen, including the lamivudine. Patients had received lamivudine for a median of 42 months and had developed HBV resistance to the drug approximately 22 months before starting therapy with adefovir dipivoxil.

Four patients withdrew from the study—two because of adverse events—leaving 31 patients who received adefovir dipivoxil for 48 weeks. Mean decreases in serum HBV-DNA concentrations from baseline were 3.40 log at week 24 and 4.01 log at week 48, similar to HBV-DNA decreases seen in HBV-monoinfected patients. Two patients underwent HBeAg seroconversion, one at week 32 and another at week 36. The drug was generally well tolerated but was associated with transient increases in ALT levels in 15 patients which, again, is similar to data from studies involving patients infected only with HBV. Renal toxicity was not seen in those with normal renal function. For those with renal dysfunction, careful monitoring and possibly dosage reductions are recommended.

Tenofovir

BASED ON PROMISING *IN VITRO* AND *IN VIVO* studies suggesting that tenofovir (Viread), Gilead Science's more successful and recently approved nucleotide analogue for the treatment of HIV, is also active against HBV, clinicians have been partial to prescribing tenofovir over adefovir for their

HIV/HBV-coinfected patients. In February 2002, preliminary data from a clinical trial evaluating tenofovir in 12 HIV/HBV-coinfected patients was reported at the 9th Conference on Retroviruses and Opportunistic Infections (Cooper, 2002). While the positive results of this one study are not enough to support tenofovir's approval for chronic hepatitis B in either HBV-monoinfected or HIV/HBV-coinfected patients, they do offer some support to clinicians opting to prescribe tenofovir over adefovir dipivoxil—at least as part of a HAART regimen intended to control both HIV and HBV replication.

Gilead Study 907 was a double-blind study that randomized patients to add either tenofovir (300 mg QD) or placebo to a background of HAART in 550 treatment-experienced patients. After 24 weeks, 14 HIV/HBV-coinfected patients were randomized to receive either tenofovir (n=12) or placebo (n=2). All of the coinfecting patients had HBV-DNA values in excess of 1 million copies/mL at baseline.

The mean decrease in serum HBV-DNA levels after 24 weeks, from baseline, was 4.63 log among the coinfecting patients who received tenofovir; a mean increase of 1.23 HBV-DNA copies/mL was observed in the coinfecting patients who received placebo. HBV-DNA responses were similar in the tenofovir treated patients, regardless of whether or not lamivudine-associated resistance mutations in HBV were documented at baseline. ALT levels normalized in three tenofovir-treated patients, and HBeAg seroconversion was documented in one patient. No new mutations were observed in HBV polymerase, indicating that tenofovir may have prolonged anti-HBV activity in both monoinfected and coinfecting patients.

Other studies looking at tenofovir in HIV/HBV-coinfected patients are currently under way. These include Gilead Study 99-903, which is assessing HBV-DNA changes in HIV/HBV treatment-naive patients assigned to receive either triple-drug therapy employing either tenofovir and lamivudine or lamivudine alone, and a study being performed by the AIDS Clinical Trials Group (A5127), a randomized study comparing adefovir dipivoxil (10 mg) to tenofovir in HIV/HBV-coinfected patients. More will be learned from the ACTG study about the comparability of the two drugs.

Famciclovir

FAMCICLOVIR, DEVELOPED BY GLAXOSMITHKLINE, is the oral form of the acyclic guanine derivative penciclovir. Final results from the first long-term multicenter placebo-controlled study of famciclovir for the treatment of HBV were published a few years ago in *Hepatology* (de Man, 2000). The study randomized patients, approximately half of whom had received previous interferon- α treatment, to receive either placebo or famciclovir (500 mg TID or 1,500 mg QD). Patients were monitored for changes in viremia, serum transaminases, and liver histology during 12 months of treatment and six months of follow-up. Accordingly, famciclovir with either dosing schedule brought about a rapid decline in HBV-DNA levels during the first eight weeks; in the placebo group, an increase in HBV-DNA was reported. As for HBeAg to anti-HBe seroconversions, none were reported in patients receiving the once-daily famciclovir, compared to 9% of those receiving the drug three times a day and 3% of those who received placebo. HBV-DNA rebounded in all patients upon completing one year of therapy. "There doesn't appear to be much of an advantage associated with famciclovir, and development of this drug for the treatment of HBV has been stopped by Glaxo-SmithKline," Dr. Peters commented. "Its efficacy is minimal, it needs to be taken three times a day if it is going to have an effect, and it has the potential to cause HBV cross-resistance to lamivudine."

Other Compounds in Development

A NUMBER OF PROMISING AGENTS CONTINUE to make their way down the HBV therapeutic pipeline. The two furthest along in development are Triangle Pharmaceuticals' emtricitabine—which is also being developed as an HIV therapeutic—and Bristol-Myers Squibb's entecavir. While emtricitabine, also known as FTC, is likely to be associated with cross-resistance to lamivudine, entecavir has shown to be effective, at least *in vitro*, against lamivudine-resistant HBV mutants (Ono-Nita, 2000).

There are also a number of compounds in phase I clinical trials. These include: several β -L-nucleosides, including Novirio Pharmaceuticals' epavudine (L-dT, NV-02B) and epcitabine (L-dC, NV-02C); Triangle Pharmaceuticals' clevudine (L-FMAU), Achillion Pharmaceuticals' β -L-Fd4C, and Eli Lilly and Company's MCC-478—all of

which are nucleoside or nucleotide analogues.

Combination Therapy

UNLIKE HIV, HBV RARELY INTEGRATES completely into the host cell genome. While integration can occur during the immune-clearance phase of chronic HBV infection, it usually occurs at random and is typically sub-genomic. In the latent phase of infection, chromosomal damage caused by these integrations, when they do occur, as well as the necroinflammatory activity associated with chronic infection combines to place the HBsAg-positive carrier at an increased risk of hepatocellular carcinoma (HCC). However, the cccDNA form—the major transcriptional template—of the viral genome persists in long-lived hepatocytes and possibly other cell types. Because HBV cccDNA is not generated or replenished directly by conventional viral DNA replication, it was believed to be unaffected by antiviral therapy. However, some recent data suggest that adefovir dipivoxil—and perhaps other nucleoside analogues—do have some effect against HBV cccDNA in hepatocytes (Werle, 2002). And, as with HIV, quiescent HBV-infected cells decline biphasically, which leads to the argument that anti-HBV therapy will need to be potent enough—and continued long enough—to snuff out any remaining HBV cccDNA. This, in turn, has some experts arguing that a combination of anti-HBV drugs might be necessary to achieve this outcome.

The results of two-drug anti-HBV combinations involving interferon- α in clinical trials have been decidedly mixed. In one international trial combining lamivudine with interferon- α in chronically infected patients who had already attempted interferon- α monotherapy, those who received the dual regimen responded similarly to those who received lamivudine monotherapy (Schiff, 1998). In another study, HBeAg seroconversions after 52 weeks were much more common in patients receiving a combination of interferon- α and lamivudine (29%), compared to those receiving either interferon monotherapy (19%) or lamivudine monotherapy (18%) (Schalm, 2000). However, as pointed out by the authors of the AASLD practice guidelines published last year in *Hepatology*, problems in the design of these two studies including sample size and limited follow-up data prevent a definitive conclusion concerning the efficacy of this particular combination (Lok, 2001). But until

further data are available, combination therapy involving interferon- α and lamivudine is not recommended.

Lamivudine and famciclovir have been studied together, based on *in vitro* and *in vivo* evidence suggesting that they have synergistic effects when combined. One clinical trial randomized 21 chronically infected patients to receive either a combination of famciclovir (500 mg TID) and lamivudine (150 mg QD) or lamivudine monotherapy (Lau, 2000). After 12 weeks of therapy, the mean antiviral efficacy—defined as a reduction in HBV-DNA viral load—was greater in the dual-nucleoside analogue group when compared to the lamivudine monotherapy group (2.5 log vs. 1.8 log decrease, respectively). Unfortunately, the follow-up time in this study was limited and, as a result, durable responses could not be accurately determined. Other drugs result in greater HBV DNA suppression, and thus famciclovir will not be further evaluated.

"The next step, logically, is to test various combinations employing pegylated interferon," Dr. Peters said, encouragingly. "There are efforts to study the pegylated interferons in combination with lamivudine, adefovir, and tenofovir. Another interesting effort is to combine antiviral agents with hepatitis B vaccination, to elicit key immune responses and potentially hasten the development of antigen-to-antibody seroconversions."

Treating HIV/HBV Coinfection

FIRST OFF, IT IS IMPORTANT THAT ALL PATIENTS diagnosed with HIV undergo serologic testing for HBV coinfection, and vice versa. Given the similar routes by which both viruses are transmitted and the high prevalence rates of HIV and HBV in certain populations, all efforts should be made to promptly diagnose—and, if necessary, treat—both infections.

In the setting of HIV infection, treatments for chronic HBV infection should be paired carefully with those being used to treat HIV. Fortunately, there is some overlap in the treatments available to treat both infections, most notably lamivudine (Epvir; Epivir-HBV), which has been approved by the U.S. Food and Drug Administration for the treatment of both infections, and tenofovir (Viread), which has shown promise in clinical trials for patients coinfecting with HIV and HBV. And considering the merits of combination

therapy for both infections, drug regimens containing “maximally suppressive” anti-HIV and anti-HBV compounds should be selected (e.g., at least one protease inhibitor or nucleoside analogue in combination with lamivudine and/or tenofovir).

Hepatotoxicity is an important issue to consider when selecting drug regimens to treat HIV and HBV in patients coinfecting with both viruses. There have been reports suggesting that nevirapine may be problematic for patients with underlying HBV infection, and there has been no shortage of data indicating that the standard dose of ritonavir (Norvir) is associated with an increased risk of hepatotoxicity in HIV-positive patients coinfecting with HBV and hepatitis C virus (HCV), although the 600 mg twice-daily dose of ritonavir is rarely employed these days. Another potential concern is hepatic steatosis, which is associated with nucleoside analogue therapy, most notably didanosine (Videx), zalcitabine (Hivid), and stavudine (Zerit).

It's also important to remember that HBV is much slower to develop resistance to lamivudine and tenofovir than HIV. Thus, continuing both drugs may still be beneficial, even when genotypic or phenotypic assays determine that HIV resistance to either of these drugs is present. There is also the risk of hepatic flares if either drug is discontinued prematurely. “It is critical that therapy with an anti-HBV drug be continued to control HBV replication, irrespective of the HIV drug-resistance profile,” Dr. Peters recommended.

Questions remain regarding the use of adefovir dipivoxil in HIV/HBV-coinfecting patients. Should this drug be used in combination with other antiretroviral agents, such as lamivudine and/or tenofovir, to treat both infections? There are few data indicating that the 10 mg dose of adefovir dipivoxil is synergistic with other antiretroviral agents used to treat HIV, and it is not known if its combination with tenofovir will increase the risk of nephrotoxicity. One possibility, then, is to use adefovir monotherapy when HBV treatment is indicated but HAART for the management of HIV is not (i.e., patients with CD4+ counts above 350 cells/mm³). While there are no data regarding the use of adefovir monotherapy in HIV/HBV-coinfecting patients, Dr. Peters said that this is a treatment option being practiced by some health-care providers.

At present, there are no data indicating

that adefovir dipivoxil induces resistance—the dose approved for the treatment of chronic HBV infection is believed to be too low to select for resistance—or cross-resistance to any of the antiretrovirals used to treat HIV. Thus, employing adefovir dipivoxil monotherapy earlier in the course of HIV infection—provided that therapy for chronic HBV is indicated—may be beneficial in terms of controlling HBV replication, with the knowledge that a powerful regimen consisting of tenofovir and/or lamivudine can be called upon when HIV therapy becomes necessary.

A Few Words on Vaccination


VACCINATION AGAINST HEPATITIS A AND B IS strongly recommended for HIV-positive people who have not been exposed to either of these viruses (see Sidebar on pages 26 and 27). However, it is important to note that vaccination success is lower in HIV-positive patients, particularly those with compromised immune systems. According to one study published in 1992, standard schedules of the HBV vaccine (20 mg at months 0, 1, and 6) induce immunity in 30% to 40% of HIV-infected patients (Bruguera, 1992). Generally speaking, patients with a CD4+ count below 200 cells/mm³ have a very poor chance of response, whereas patients with counts of more than 500 CD4+ cells/mm³ can achieve antibody response in up to 70% of cases.

One study team has tested the hypothesis that doubling the number of hepatitis B vaccine injections might increase the anti-HBs response rate in HIV-positive patients (Rey, 2000). Twenty-eight HIV-infected patients with CD4+ counts greater than 200 cells/mm³, all of whom were on stable HAART and had never been vaccinated against HBV, were given three intramuscular injections of Genhevac B (20 mg) at one-month intervals. Initial nonresponders were given three additional monthly injections.

The response rate after three 20 mg injections was 55%, with the response rate being lowest (33%) among patients with CD4+ counts between 200 and 500 cells/mm³, compared to a higher response rate (87.5%) among patients with CD4+ counts above 500 cells/mm³. Among nine initial nonresponders, only two failed to respond to three additional doses; thus, the overall response rate was 90%. One year later, only 10/17 (58.8%) evaluable pa-

tients had protective anti-HBs. While the study authors concluded that doubling the number of hepatitis B vaccinations in HIV-infected patients might significantly improve anti-HBs response rate, it is not likely to be recommended, given its short-lived persistence.

Conclusion

IN SUMMARIZING HER TALK, DR. PETERS reiterated some of the more important recommendations regarding the diagnosis and care of HIV-patients coinfecting with HBV. “As a part of an initial workup of an HIV-infected patient, diagnostic serologies should be performed, including HBsAg and anti-HBc. If these are positive, serum HBV-DNA should be quantified and monitored regularly, along with alpha-fetoprotein levels and imaging studies of the liver.” Regarding the natural history of chronic HBV in HIV-infected patients, Dr. Peters reminded PRN members that the immune response to HBV predicts disease outcome and that while immunodeficiency may decrease necroinflammation, it allows for increased replication of HBV. “The immune recovery promoted by HAART allows for necroinflammation to continue. There's no doubting the importance of treating HBV. Treatment should be considered where there is HBV viremia and elevated transaminases. And remember, HBV should be treated along with HIV and combination regimens should involve antiviral compounds active against both viral infections.” 

References

- Benhamou Y, Bochet M, Thibault V, et al. **Safety and efficacy of adefovir dipivoxil in patients co-infected with HIV-1 and lamivudine-resistant hepatitis B virus: an open-label pilot study.** *Lancet* 358:718-23, 2001.
- Benhamou Y, Bochet M, Thibault V, et al. **Long-term incidence of hepatitis B virus resistance to lamivudine in human immunodeficiency virus-infected patients.** *Hepatology* 30(5):1302-6, 1999.
- Bruguera M, Cremades M, Salinas R, et al. **Impaired response to recombinant hepatitis B vaccine in HIV-infected persons.** *J Clin Gastroenterol* 14:27-30, 1992.
- Cooksley WGE, Piratvisuth, T, Wang Y-J, et al. **Evidence for the efficacy of Peg-Interferon alfa 2A (40Kd) (Pegasys) in the treatment of HBeAntigen positive chronic hepatitis B and impact of baseline factors.** 37th Annual Meeting of the European Association for the Study of the Liver, Madrid, 2002.

TABLE 1. A Review of the Interpretation of Hepatitis B Serologic Results							
Stage of infection	Anti-HBs	HBsAg	Anti-HBc (IgG)	Anti-HBc (IgM)	HBeAg	Anti-HBe	HBV-DNA
Acute (early)	-	+	+	+	+	-	+
Acute (resolving)	-	+	+	+	-	+/-	-
Chronic (high infectivity)	-	+	+	-	+	-	+
Chronic (high infectivity); precore mutant	-	+	+	-	-	+/-	+
Chronic; abnormal serologies (possibility in HIV infection)	-	-	+	-	+/-	+/-	+
Chronic (low infectivity)	-	+	+	-	-	+/-	-
Resolved (latent)	+	-	+	-	-	+/-	-
Successful vaccination	+	-	-	-	-	-	-

Source: Based on HBV serology interpretations provided by Marion Peters, MD; the U.S. Centers for Disease Control and Prevention (Atlanta); the Association for Genitourinary Medicine (London); and the Medical Society for the Study of Venereal Disease (London).

Cooper DA, Cheng A, Coakley WGE, et al. **Anti-HBV Activity of Tenofovir Disoproxil Fumarate (TDF) in Lamivudine (LAM) Experienced HIV/HBV Co-Infected** [Abstract 124]. 9th Conference on Retroviruses and Opportunistic Infections, Seattle, 2002.

de Man RA, Macellin P, Habal F, et al. **A randomized, placebo-controlled trial to evaluate the efficacy of twelve-month famciclovir treatment in patients with chronic HBeAg-positive hepatitis B.** *Hepatology* 32:413-7, 2000.

Dienstag JL, Schiff ER, Wright TL, et al. **Lamivudine as initial treatment for chronic hepatitis B in the United States.** *N Engl J Med* 341:1256-63, 1999.

Gilson RJ, Chopra K, Murray-Lyon IM, et al. **A placebo-controlled phase I/II study of adefovir dipivoxil (bis-POM PME) in patients with chronic hepatitis B infection.** *Hepatology* 24:281A, 1996.

Hadziyannis S, Tassopoulos N, Heathcote E, et al. **GS-98-438 a double-blind, randomized, placebo-controlled study of adefovir dipivoxil (ADV) for presumed precore mutant chronic hepatitis B: 48 week results** [Abstract 648]. 37th Annual Meeting of the European Association for the Study of the Liver, Madrid, 2002.

Heathcote E, Jeffers L, Perrillo R, et al. **Sustained antiviral response and lack of viral resistance with long term adefovir dipivoxil (ADV) therapy in chronic HBV infection** [Abstract 590]. 37th Annual Meeting of the European Association for the Study of the Liver, Madrid, 2002.

Heathcote EJ, Jeffers L, Wright T, et al. **Loss of serum HBV DNA and HBeAg and seroconversion following short term (12 weeks) adefovir dipivoxil therapy in chronic hepatitis B: two placebo-controlled phase II studies.** Adefovir Dipivoxil HBV Study Team. *Hepatology* 28:317A, 1998.

Hofer M, Joller-Jemelka HI, Grob PJ, et al. **Frequent chronic hepatitis B virus infection in HIV-infected patients positive for antibody to hepatitis B core antigen only.** *Swiss HIV Cohort Study.* *Eur J Clin Microbiol Infect Dis* 17(1):6-13, 1998.

Jeffers J, Heathcote E, Wright T, et al. **A phase II dose-ranging, placebo-controlled trial of adefovir dipivoxil for the treatment of chronic hepatitis B virus infection.** *Antiviral Res* 37:A197, 1998.

Lai CL. **Experience with lamivudine in Asian Trials. Hepatitis B 2000: Therapeutic Update.** Satellite Symposium at the 10th International Symposium on Viral Hepatitis and Liver Disease, Atlanta, 2000.

Lai CL, Chien RN, Leung NW, et al. **A one-year trial of lamivudine for chronic hepatitis B.** Asia Hepatitis Lamivudine Study Group. *N Engl J Med* 339:61-8, 1998.

Lau GK, Tsiang M, Yuen ST, et al. **Combination therapy with lamivudine and famciclovir for chronic hepatitis B-infected Chinese patients: a viral dynamics study.** *Hepatology* 32:394-9, 2000.

Leung NW, Lai CL, Chang TT, et al. **Extended lamivudine treatment in patients with chronic hepatitis B enhances hepatitis B e antigen seroconversion rates: results after 3 years of therapy.** *Hepatology* 33(6):1527-32, 2001.

Lok AS, McMahon BJ. **Chronic hepatitis B.** *Hepatology* 34(6):1225-41, 2001.

MacDonald J, Caruso L, Karayannis P, et al. **Diminished responsiveness of male homosexual hepatitis B virus carriers with HTLV III antibodies to recombinant alpha interferon.** *Hepatology* 7:719-23, 1987.

Marcellin P, Chang T-T, Lim SG, et al. **A double blind, randomized, placebo-controlled study of adefovir dipivoxil (ADV) for the treatment of patients with HBeAg+ chronic hepatitis B infection: 48 week results (GS-98-437)** [Abstract 674]. 52nd Annual Meeting of the American Association for the Study of Liver Diseases, Dallas, 2001.

Ono-Nita SK, Kato N, Shiratori Y, et al. **Influence of B domain mutation (L528M) of the hepatitis B virus polymerase on replication ability and resistance to nucleoside analogues.** *Hepatology* 32:393A, 2000.

Rey D, Krantz V, Partisani M, et al. **Increasing the number of hepatitis B vaccine injections augments anti-HBs response rate in HIV-infected patients. Effects on HIV-1 viral load.** *Vaccine* 18(13):1161-5, 2000.

Schalm SW, Heathcote J, Cianciara J, et al. **Lamivudine and alpha interferon combination treatment of patients with chronic hepatitis B infection: a randomised trial.** *Gut* 46(4):562-8, 2000.

Schiff E, Karayalcin S, Grimm I, et al. **A placebo controlled study of lamivudine and interferon alpha-2b in patients with chronic hepatitis B who previously failed interferon therapy.** International Lamivudine Investigator Group. *Hepatology* 28:388A, 1998.

Tassopoulos NC, Volpes R, Pastore G, et al. **Efficacy of lamivudine in patients with hepatitis B e antigen-negative/hepatitis B virus positive (precore mutant) chronic hepatitis B.** Lamivudine Precore Mutant Study Group. *Hepatology* 29:889-96, 1999.

Thio CL, Seaberg EC, Skolasky R, et al. **Liver disease mortality in HIV-HBV coinfecting persons** [Abstract 656]. 9th Conference on Retroviruses and Opportunistic Infections, Seattle, 2002.

Werle B, Wursthorn K, Petersen J, et al. **Development of a quantitative assay for hepatitis B CCC DNA levels in patients with chronic hepatitis B** [Abstract 638]. 37th Annual Meeting of the European Association for the Study of the Liver, Madrid, 2002.

Wong D, Yim C, Naylor C, et al. **Interferon alfa treatment of chronic hepatitis B: Randomized trial in a predominantly homosexual male population.** *Gastroenterology* 108:165-71, 1995.

Yang H, Westland C, Delaney W, et al. **Lack of emerging resistance mutations in 467 HBeAg- and HBeAg+ patients with chronic hepatitis B receiving adefovir dipivoxil for 48 weeks** [Abstract 577]. 37th Annual Meeting of the European Association for the Study of the Liver, Madrid, 2002.