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Management and treatment of chronic hepatitis B virus : Belgian Association for the Study of the Liver (BASL) 2007 guidelines

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I. Introduction

Chronic hepatitis B virus (HBV) infection currently affects about 400 million people and is responsible for 500,000 to 1,000,000 deaths annually worldwide from cirrhosis and hepatocellular carcinoma (HCC) (1). For this reason, screening high risk populations to identify HBV infected persons is important so that guidelines for treatment and prevention of transmission can be given in this specific group.

Recently, new drugs became available for HBV and new insights in resistance and definitions came up. So, the purpose of this paper is providing an update of the recent literature and guidelines concerning

1. screening for chronic hepatitis B (CHB)
2. management of patients with CHB
3. treatment of CHB in mono-infected patients and in special patient populations (co-infected, transplanted and immunosuppressed patients).

The recommendations are based on published information and the level of evidence is reported with each recommendation. The level of evidence is graded as : grade I : randomized controlled trials ; grade II-1 : controlled trials without randomization ; grade II-2 : cohort or case-control analytic study ; grade II-3 : multiple time series, dramatic uncontrolled experiments ; grade III : descriptive epidemiology, expert opinions.

II. Natural history of hepatitis B

II.1. *Characterization of different populations with chronic HBsAg positivity*

Both the severity of acute hepatitis and the risk of developing chronic hepatitis B and of suffering from the consequences of this chronic infection are related to the age at which the infection is acquired. So there are three different patterns of patients suffering from chronic hepatitis B infection (presence of HBsAg more than 6 months). In Table 1 the different profiles of chronic hepatitis B are summarized.

The **first pattern** is seen in individuals in Western developed countries. The hepatitis B virus is acquired during adulthood and transmission is via sexual exposure or intravenous drug use. After acute infection (which can be severe), the risk of chronicity is low (10%). When chronicity occurs, the early phase of chronic infection is marked by the continued presence of HBsAg, high levels of serum HBV DNA, the presence of hepatitis Be antigen (HBeAg), and increased amino alanine aminotransferase (ALT) levels. The duration of this *HBeAg-positive chronic hepatitis* can be prolonged and severe and may result in cirrhosis. For many persons, however, it does not cause clinical symptoms. HBeAg seroconversion (defined as loss of HBeAg and acquisition of anti-HBe antibody) occurs spontaneously (overall, the estimated annual rate of spontaneous seroconversion is 8-12%). The seroconversion is preceded by a marked decrease in serum HBV DNA levels to less than 20,000 IU/mL and is typically followed by normalization of ALT levels (sometimes preceded by a transient flare of disease with marked elevation of ALT levels). This second phase is called *inactive HBsAg carrier state*. The inactive carrier state generally (60-80%) takes a benign course, but can be reactivated either spontaneously or by immune suppression.

A proportion of patients (up to one-third) who undergo HBeAg seroconversion have a return of high levels of HBV DNA and persistent (30-40%) or intermittent (45-65%) increases of ALT levels. Most of these patients have a naturally occurring mutant form of HBV that abolishes or down-regulates HBeAg production, usually because of *mutation in the precore or core promoter region*. This form of chronic HBV infection is called *HBeAg-negative chronic hepatitis B*. This form can

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Table 1. — **Chronic hepatitis B profiles**

	Inactive carrier	Immune tolerant phase	Active chronic HBV wild type	Active chronic HBV precore mutant
HBsAg	+	+	+	+
HBeAg	-	+	+	-
Anti HBe Ab	+	-	-	+
ALT	normal	normal	elevated	elevated
HBV DNA Copies/ml	$\leq 10^4$	$> 10^{5-7}$	$> 10^5$	10^4-10^5
HBV DNA IU/ml	≤≤ 2,000	> 20,000	> 20,000	2,000-20,000

Abbreviations : HBsAg : hepatitis Bs antigen ; HBeAg : hepatitis Be antigen. anti HBe Ab : anti HBe Ab hepatitis Be antibodies ; ALT : alanine aminotransferase.

occur either close to HBeAg seroconversion or many years or even decades later. HBV DNA levels tend to be elevated, but are lower, however, compared with those observed in the HBeAg-positive variant. The consequences of HBeAg-negative chronic hepatitis can be as severe as the HBeAg-positive chronic hepatitis B variant. Therefore, serial HBV DNA testing is necessary to determine if a HBsAg-positive, HBeAg-negative carrier is truly in the inactive carrier state and life-long follow-up is required to confirm that the inactive state is maintained.

The <u>*second pattern*</u> is seen most often in Asia, where the majority of chronic HBV infections result from perinatal transmission. Acute hepatitis in infants is typically asymptomatic, but chronicity occurs in more than 90% of cases. Perinatally acquired HBV infection is associated with immunologic tolerance to the virus (*immune tolerant phase*) and despite high levels of HBV DNA, ALT levels remain in the normal range. This phase may last decades, with low rates of spontaneous HBeAg seroconversion. Most studies suggest that very little injury occurs during this phase. The disease activity can accelerate in some of these patients, rarely before adulthood, with increased ALT levels and typical HBeAg-positive chronic hepatitis B *(immune clearance phase)*. The transition to chronic hepatitis B can be abrupt and resemble acute hepatitis with a transaminase flare. The HBeAg seroconversion occurs in the majority of patients only after the third or fourth decade of infection, after which the patients can enter the state of *inactive carriership*. However, those Asian patients who clear HBeAg may harbour precore and core mutants eventually leading to *HBeAg-negative chronic hepatitis B.*

The <u>*third pattern*</u> is seen in Africa and Mediterranean countries, where transmission tends to be person-to-person during childhood. Children exposed to the virus within the first 5 years of life have a 25-50% risk of developing chronic infection. Most HBeAg-positive children have elevated ALT levels, and seroconversion to anti-HBe is common near or shortly after the onset of puberty. The natural history of this population is intermediate between that of Western and Asian populations.

Some patients (approximately 0.5% per year) also lose HBsAg and develop anti-HBs, which is referred to as *resolution of HBV infection*. The prognosis is improved in these patients. However, low levels of HBV DNA remain detectable in serum in up to half of these persons and reactivation is possible in case of immunosuppression or chemotherapy. Furthermore, hepatocellular carcinoma has been reported years after clearance of HBsAg, particularly in those who had progressed to cirrhosis or had a highly active disease before HBsAg clearance.

II.2. *Role of HBV genotypes in the evolution of chronic HBV*

Besides the age at which the infection is acquired, some of the variations in outcome of HBV infection may be related to the genetic heterogeneity of the virus. Eight genotypes (A-H) have been described, genotype A being most common in the United States and Northern Europe, B and C in Asia, and D in Mediterranean countries and the Middle East. Chronic infection with genotype B appears to have a better prognosis than genotype C. Precore mutant infection is also most common in genotypes B, C and D, which explains why precore mutant infection is more common in Asia and Southern Europe (2).

II.3. *Role of HBV DNA levels in evolution of chronic HBV*

There is a relationship between serum HBV DNA level and prognosis : the cumulative incidence of cirrhosis and hepatocellular carcinoma (HCC) is 4.5% and 1.3%, respectively, in persons with DNA levels less than 300 copies/mL (≈ 50 IU/mL), and 36.2% and 14.9%, respectively, in persons with DNA levels of $\geq 10^6$ copies/mL (≈ 200,000 IU/mL). This is the rationale for treating patients with high levels of HBV DNA.

A high level of ALT is the most important predictive factor of response to interferon and nucleo(t)side antiviral therapies. This is the rationale for considering therapies mainly for patients with elevated ALT levels. However, treatment is sometimes necessary in patients

with normal liver tests but with important fibrosis or inflammation.

Patterns of infection are not *per se* predictors of response to therapies : Asian patients with increased ALT levels respond to interferon and lamivudine as well as Caucasian patients. Many Asian patients, however, have normal ALT levels at presentation and thus lower response rates to therapy.

III. Identification of infected persons, screening and vaccination

III.1. *Prevalence*

The prevalence of HBsAg varies greatly from country to country : some (including Belgium) having a low prevalence (< 2%), some having intermediate (2%-8%) or high prevalence (≥ 8%) of HBV infection (3-6). In developed countries, the prevalence is higher among immigrants from high or intermediate prevalence countries and in those with high risk behaviours (3,5).

III.2. *Screening HBV*

The *screening tests for chronic HBV* should include HBsAg and hepatitis B surface antibody (anti-HBs). Hepatitis B core antibody (anti-HBc) can be used as a marker of contact with HBV : this can be an immune marker after infection, chronic hepatitis B, acute HBV (anti HBc IgM+) in window phase. Thus positivity for HBcAb should be followed by testing for both HBsAg and anti-HBs to differentiate infection from immunity (7).

III.3. *Hepatitis B vaccination*

In the eighties recombinant DNA technology enabled the expression of HBsAg in other organisms ; as a result, several recombinant DNA vaccines against hepatitis B from different manufacturers have become available since 1986. In most hepatitis B vaccines, aluminium salts are used to adsorb the HBsAg and to enhance the immune response (8).

A series of (intramuscular) vaccinations are required to induce immunity against HBV. Following a full course of vaccination, seroprotection rates (anti-HBs ≥ 10 IU/L) for antibodies against hepatitis B surface antigen (anti-HBs) is close to 100% in children and almost 95% in healthy adults. A large variety of hepatitis B vaccination schedules have been shown to induce seroprotective anti-HBs levels in healthy infants and children : these include schedules where doses are administered at birth, 1 and 6 months of age ; at 2, 4, and 6 months of age ; and 6, 10, and 14 weeks of age. A full course of vaccination in adolescents and adults comprises a 0, 1, and 6 month vaccine administration or a 0, 1, 2 and 12 month schedule (9-15).

People who are elderly, obese, heavy smokers, or immunocompromised, including those infected with HIV, may have suboptimal responses when vaccinated. Immunodeficient patients, such as those undergoing hemodialysis or immunosuppressant therapy, require higher doses of vaccine and more injections (at months 0, 1, 2, and 6) to achieve an adequate immune response. Follow-up studies have shown that vaccine-induced antibody persists over periods of at least 10 to 15 years and that duration of anti-HBs is related to the antibody peak level achieved after primary vaccination. Follow-up of vaccinees has shown that the antibody concentrations usually decline (and may disappear) over time, but clinically significant breakthrough infections are rare in successfully vaccinated people (achieving anti-HBs ≥ 10 IU/L, when measured 1-3 months after a completely administered hepatitis B vaccination course) (16). Evidence indicates that successfully vaccinated individuals who have lost antibody over time usually show a rapid anamnestic response when boosted with an additional dose of vaccine given several years after the primary course of vaccination or when exposed to the HBV. This means that the immunological memory for HBsAg can outlast the anti-HBs detection, providing long-term protection against acute disease and the development of the HBsAg carrier state (16). Hence, for immunocompetent children and adults the routine administration of booster doses of vaccine does not appear necessary to sustain long-term protection. Such conclusions are based on data collected during the first 10 to 20 years of vaccination in countries of both high and low endemicity. However, additional long-term follow-up and surveillance in hepatitis B vaccinees in different countries are still warranted in order to establish whether a primary course of vaccination of healthy individuals can further confer lifelong protection without a need for additional booster injections (17-23).

Recommendations for which persons should be tested for hepatitis B (7) (evidence level I)

– Persons born in hyperendemic areas ; men who have sex with men ; persons who have at any time injected drugs ; renal dialysis patients ; HIV and/or HCV-infected individuals ; all pregnant women, family members, household members and any sexual contacts of HBV-infected persons ; promiscuous persons or those with a history of sexually transmitted diseases, prisoners or past-prisoners ; persons with chronically elevated ALT or AST levels.

– Those high risk patients who are HBsAg negative should be recommended for vaccination (24).

Recommendations for counseling and prevention of transmission of HBV (7)

– All HBV carriers should be informed about the possible transmission pathways of HBV and take certain measures : use a barrier method during intercourse if the partner is not immune, avoid blood contact with other persons (do not share razors and toothbrush, cover wounds and scratches, do not donate blood, sperm

or organs, clean blood with detergents), household and sexual contacts who are negative for HBV markers should be vaccinated for HBV, newborns of infected mothers should receive within 12-24 h after delivery hepatitis B immune globulins (HBIg) and hepatitis B vaccination, followed by a complete vaccination scheme (24) (evidence level I, III).
– Vaccination for HBV is also recommended in high risk populations such as health care workers, renal dialysis patients and children of HBsAg positive mothers (7, 24) *(evidence level II-2).*

IV. Effect of treatment on natural history and cost-effectiveness of treatment

IV.1. *Effect of treatment on natural history*

The course of chronic hepatitis B is mostly asymptomatic until cirrhosis and/or HCC (which may occur in the absence of cirrhosis) arise. As a consequence, the major goal of therapy is to prevent disease, development of cirrhosis and its complications including HCC. All anti-HBV drugs showed evidence of treatment benefit using biochemical, virological and histological endpoints. Biochemical and histological improvements also occurred more frequently after antiviral therapy than with placebo.

A randomized controlled trial from Taiwan, showed that long-term lamivudine therapy is associated with improved patient survival, decreased incidence of hepatic decompensation and lower rates of HCC in patients with cirrhosis (25). The benefits were mostly seen in those who maintained a virological response and those who did not develop lamivudine resistance. On the other hand, the presence of high HBV DNA levels is associated with increased disease progression to decompensated cirrhosis, need for liver transplantation and the development of HCC (26,27). As a consequence it seems indicated that patients with advanced fibrosis and cirrhosis and high levels of HBV DNA should receive therapy.

IV.2. *Cost-effectiveness of HBV treatment*

The costs of therapy for chronic hepatitis B are high and should be considered when initiating treatment. In Table 2 the costs of each treatment available in Belgium and the US is shown. However, additional costs for therapeutic monitoring, the impact of emerging resistances, and the disease progression should all be taken into account when looking at cost-effectiveness.

There are only a few studies that have looked at the cost effectiveness of hepatitis B treatment. Although lamivudine is associated with a high risk of resistance, many patients receive initial therapy with lamivudine (Zeffix®), in part because this agent is less expensive than the more recently approved adefovir (Hepsera®) and entecavir (Baraclude®). However, a study (using a mathematical model) showed that treatment with entecavir is cost-effective compared with either lamivudine or adefovir (28). Both HBeAg seroconversion and HBV DNA suppression were markers for treatment success (28). The use of entecavir was associated with an increase in life expectancy and a decrease in incremental cost effectiveness ratio (28). Two studies demonstrated the cost-effectiveness of adefovir treatment (29,30).

Table 2. — **Costs of therapy in Belgium and US (for entecavir, telbivudine, tenofovir and emtricitabine)**

Drug	Cost per year
Peg-interferon alpha 2a	11,468 EUR
Lamivudine	749 EUR
Adefovir	6,159 EUR
Tenofovir	5,811 USD*
Entecavir	6,159 EUR °°
Telbivudine	4,536 EUR ° or 5,924 USD*
Emtricitabine	3,872 USD*

* To date there are no prices available for these drugs in Belgium ; we used the prices from the paper of Hoofnagle *et al.* (41).
° : the prices given for telbivudine are those of our surrounding European countries, the price in Belgium has not yet been published.
°° : the price for entecavir 1mg and not for the 0.5 mg dose.

Two studies looked at the hepatitis B management costs in Europe (France, Italy, Spain and UK) and in Canada (31,32). Both studies came to the same conclusions : the average yearly cost per patient by liver disease state increased with increasing disease progression, culminating in the highest cost, liver transplantation. The estimated cost in year 2001 was as follows : for chronic hepatitis B : € 1,093 – € 3,396 ; for compensated cirrhosis : € 1,134 – € 3,997 ; for decompensated cirrhosis : € 5,292 – € 8,842 ; for hepatocellular carcinoma : € 3,731 – € 9,352 and for liver transplantation : € 25,165 – € 84,568 (31).

With the high costs associated with the disease, it is absolutely necessary to prevent hepatitis B by promoting HBV vaccination programs. Any effective treatment that stops disease progression or even better reverses the disease state will be economically beneficial (31,32).

V. Indications for treatment in chronic hepatitis B (CHB)

V.1. *Goals for therapy in CHB*

The main goal of therapies is to significantly suppress HBV replication, thus preventing liver disease progression to cirrhosis and its complications including hepatocellular carcinoma.

In patients who are HBeAg-positive, the treatment goal is HBeAg seroconversion (anti-HBe Ab occurrence) with sustained suppression of HBV DNA and hopefully HBsAg seroconversion.

In patients who are HBeAg-negative, the therapeutic goal is sustained suppressed HBV DNA and HBsAg seroconversion (anti-HBs Ab occurrence).

There exist *4 types of response* : biochemical response (ALT levels decrease within normal range) ;

virological response (HBV DNA decreases to undetectable levels by PCR and loss of HBeAg in patients who were HBeAg positive at start) ; histologic response (decrease with at least 2 points in the histology activity index and no worsening in fibrosis compared with pre-treatment biopsy) and complete response (biochemical and virological response with loss of HBsAg) (7). Primary non-response is currently defined (only for patients treated with nucleos(t)ide analogues) as a decrease in HBV DNA with < 1 $\log_{10}$ IU/mL after 6 months of treatment (33). Virologic relapse means an increase in HBV DNA of > 1 $\log_{10}$ IU/mL after treatment discontinuation in 2 subsequent blood samples 1 month apart in patients who have responded and have been compliant with the antiviral medication (7,33).

V.2. *Standardization of HBV DNA levels*

Because of the different methods used to quantify HBV DNA levels, significant variations are observed in viral load results. In order to enable standardization of viral load results and to allow comparison between studies, the World Health Organization (WHO) has established an international reference standard for HBV DNA units, set at IU/mL (1 IU/mL ≈ 5.6 copies/mL) (34). So, 10^4 copies/mL ≈ 2000 IU/mL ; 10^5 copies/mL ≈ 20,000 IU/mL ; 10^6 copies/mL ≈ 200,000 IU/mL etc.

V.3. *Candidates for therapy* (7,35)

Recommendations for treatment indications

HBeAg-positive patients (Fig. 1)

- *Patients with high levels of DNA (> 20,000 IU/mL) and elevated ALT levels greater than twice the upper limit of normal are the most appropriate candidates for therapy (evidence level I).*
- *Patients with high levels of HBV DNA (> 20,000 IU/mL) with persistent borderline normal or slightly elevated ALT (immune tolerant phase), have a low probability of obtaining HBeAg sero-*

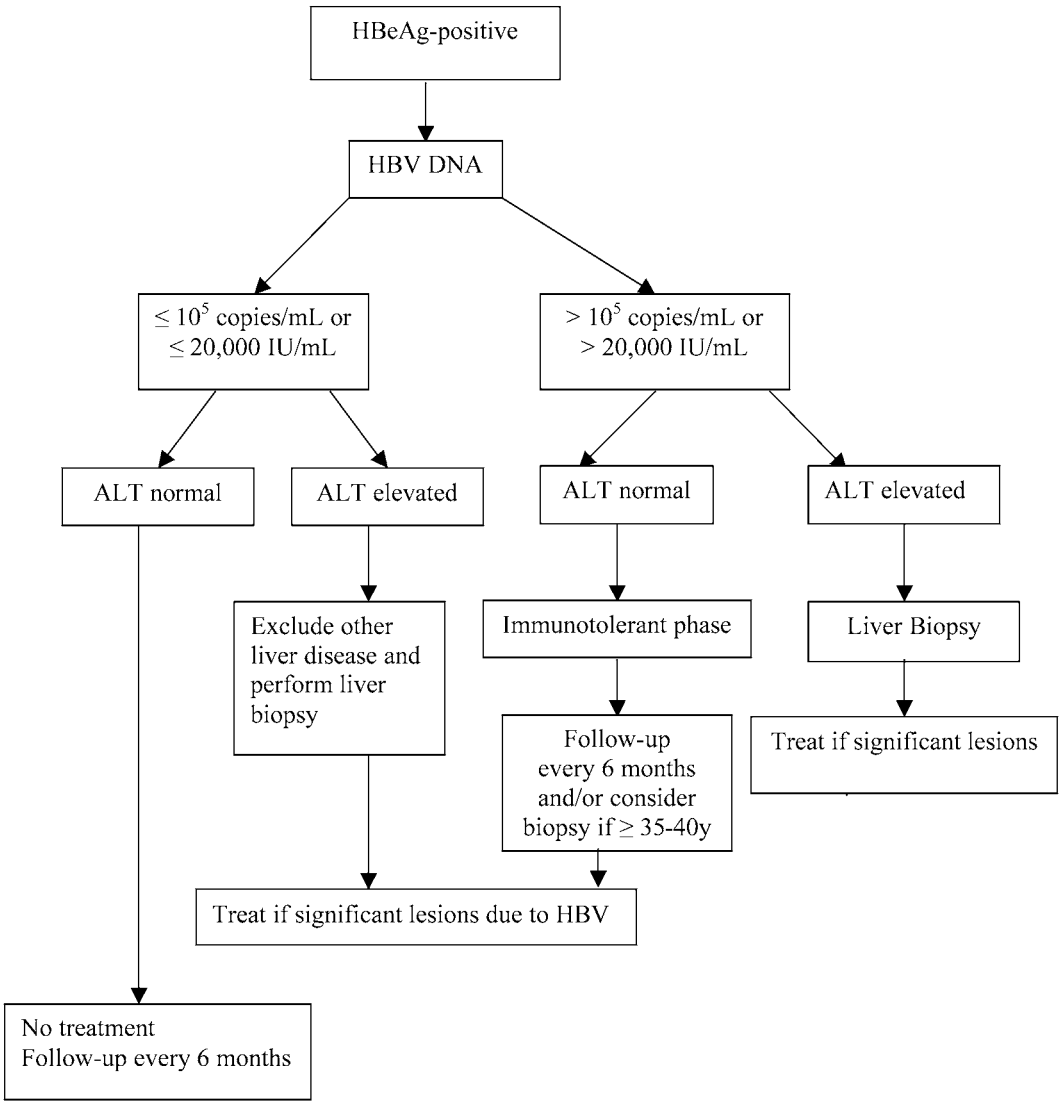

Fig. 1. — Management algorithm for patients with HBeAg-positive chronic HBV

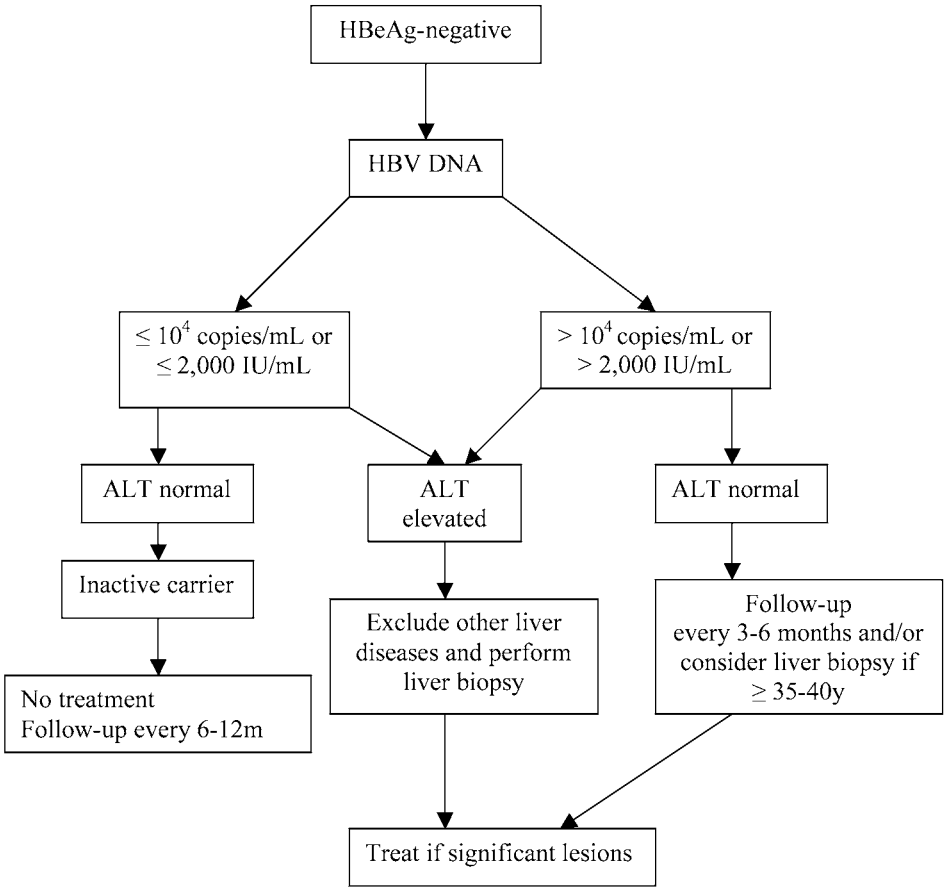

Fig. 2. — Management algorithm for patients with HBeAg-negative chronic HBV

conversion with treatment. Liver biopsy should be considered, particularly in individuals older than 35-40 years of age, and treatment should be considered if moderate/severe inflammation or significant fibrosis are found. Liver biopsy is usually not necessary in young patients (below 30 years of age) who are HBeAg-positive and have persistently normal ALT. In the absence of biopsy, patients should be monitored to observe for increase in ALT levels (evidence I, II-3).

- *Patients with low levels of HBV DNA (≤ 20,000 IU/mL) and normal ALT are not routinely recommended for treatment. Due to the low levels of HBV DNA, the majority of these individuals are at low risk of disease. They should be monitored to ensure stability of HBV DNA and ALT levels (evidence level I).*
- *Patients with low levels of HBV DNA (≤ 20,000 IU/mL) and elevated ALT levels should have liver biopsy and other causes of liver diseases should be excluded (evidence level II-3).*

HBeAg-negative patients (Fig. 2)

The recommendations are the same as for HBeAg-positive patients. The threshold of HBV DNA levels for considering a treatment is however lower : > 2,000 IU/mL (evidence level I, II-1, II-2)

In decompensated patients :

Treatment is considered in the presence of detectable HBV DNA, regardless the HBeAg status (evidence level II-1).

VI. Treatment options

The primary aims in the treatment of patients chronically infected with HBV are achieving sustained suppression of HBV replication and preventing cirrhosis, hepatocellular carcinoma and liver failure.

Currently, 5 licensed therapies are available in Europe and 6 in the United States (US) :

Nucleos(t)ide Analogues (NA) : lamivudine (Zeffix®), adefovir dipivoxil (Hepsera®), entecavir (Baraclude®), telbivudine (Sebivo®) and interferon-based therapy with interferon α_{2a} (Roferon A®) and interferon α_{2b} (Intron A®), pegylated interferon-α_{2a} (Pegasys®) (Peg-IFN-α_{2a}).

Nucleos(t)ide analogues are chain terminators that block the HBV polymerase and hence viral replication. They often need to be administered for prolonged periods and often indefinitely. Interferon based therapy enhances the immune clearance of the virus and is given for a fixed time period.

We will discuss below the suggested treatment options for different patient populations.

VI.1. *Interferon and pegylated interferon in HBeAg + and HBeAg – patients*

Pegylated interferon-α_2 (Peg-IFN-$_{2a}$)

Peg-IFN-α_{2a} (40kD)(Pegasys®) is a recombinant interferon-α_{2a} (IFN-α_{2a}) covalently bound to a 40 kD branched polyethylene glycol (Peg) molecule. Pegylation increases systemic exposure by decreasing clearance of the molecule, resulting in a prolonged drug effect. Peg-IFN has both immunomodulatory effects and inhibits viral replication and /or viral functions (36). In this review we will only discuss the use of Peg-IFN-α_{2a} as it is probably more efficacious than IFN-α_2 and its use is easier : 1 subcutaneous (SC) administration per week (Peg-IFN-α_{2a}) versus three times per week or daily SC injections for the IFN-α_{2a} (37).

Peg-IFN-α_{2b} (12kD)(PegIntron®) is a recombinant interferon-α_{2b} (IFN-α_{2b}) covalently bound to a 12kD branched polyethylene glycol (Peg) molecule. However, currently only Peg-IFN-α_{2a} is licensed for treatment of hepatitis B.

VI.1.1. HBeAg-positive chronic hepatitis B

In a phase III study, patients were randomised in 3 arms : Peg-IFN-α_{2a} + placebo or Peg-IFN-α_{2a} + lamivudine 100 mg/d orally (po) or lamivudine 100 mg/d po alone (38). Despite the more important drop in HBV DNA at the end of treatment in the combined Peg-IFN-α_{2a} + lamivudine arm (- 7.2 log) compared to Peg-IFN-α_{2a} alone (-4.5 log) or lamivudine alone (-5.8 log), this did not result in more HBeAg or HBsAg seroconversions. HBeAg seroconversion 6 months after the end of treatment occurred in 32%, 27% and 19% in respectively Peg-IFN-α_{2a} alone, Peg-IFN-α_{2a} + lamivudine and lamivudine monotherapy ($p < 0.001$ for Peg-IFN-α_{2a} alone versus lamivudine ; $p < 0.02$ for Peg-IFN-α_{2a} + lamivudine versus lamivudine) (38). HBsAg seroconversion was observed in 3%, 3% and 0% respectively (38). Similar results were reported with Peg-IFN-α_{2b} (39,40). Histological response rates were similar in the 3 groups : 38%, 41% and 34% respectively (38).

Currently, peg-IFN-α_{2a} appears to be superior to nucleos(t)ide analogues, especially due to the highest rates of HBeAg and HBsAg loss after 1 year treatment and the absence of resistance. Thus Peg-IFN-α_{2a} monotherapy can be proposed as a first-line therapy in HBeAg-positive patients (7,35,41).

Combination with lamivudine does not lead to a better viral response (7,38,41). Combination therapy Peg-IFN-α_{2a} with adefovir can have favourable effects on cccDNA in the liver, however, results are too scarce to give guidelines.

VI.1.2. HBeAg-negative chronic hepatitis

In a phase III study, patients were randomised in 3 arms : Peg-IFN-α_{2a} + placebo or Peg-IFN-α_{2a} + lamivudine 100 mg/d orally (po) or lamivudine 100 mg/d po alone (42). Despite the more important drop in HBV DNA at the end of treatment in the combined Peg-IFN-α_{2a} + lamivudine arm (- 5 log) compared to Peg-IFN-α_{2a} alone (-4.1 log) or lamivudine alone (-4.2 log), this did not result in more sustained viral response or HBsAg seroconversions. HBV DNA below 400 copies/mL 6 months after the end of treatment occurred in 19%, 20% and 7% in respectively Peg-IFN-α_{2a} alone, Peg-IFN-α_{2a} + lamivudine and lamivudine monotherapy ($p < 0.001$ for both comparisons with lamivudine monotherapy) (42). HBsAg seroconversion was observed in 4%, 3% and 0% respectively (42). Histological response rates were similar in the 3 groups : 48%, 38% and 40% respectively (42).

Three years post treatment with Peg-IFN-α_{2a} during 48 weeks leads to a sustained normalisation of ALT in 31% and HBV DNA levels below 10,000 copies/mL and below 400 copies/ml in 28% and 18% of the patients with HBeAg-negative chronic hepatitis B. The number of patients losing HBsAg increased over time to 8% at 3 years post-treatment (43)

Thus Peg-IFN-α_{2a} can be used as first-line therapy for chronic HBeAg-negative patients (7,35,41).
We should notice that in these trials of Marcellin et al and Lau et al, withdrawal of lamivudine was associated with transient exacerbation of the disease and led in some cases to mortality (41,44).

VI.1.3. Dosage and administration of Peg-IFN-α_{2a}

The recommended dosage of Peg-IFN-α_{2a} is 180 µg administered once weekly by subcutaneous (SC) injection for 48 weeks. Dose reduction down to 135 µg/week SC in patients with end stage renal disease undergoing haemodialysis is recommended (36).

VI.1.4. Predictors of Response to Peg-IFN-α_{2a}

Predictors of response to Peg- IFN-α_{2a} treatment in HBeAg-positive patients are (40,41,45,46) :
- high baseline ALT levels [> twice normal value (> 2N), or even better > 5N]
- low baseline viral HBV DNA load (< 7 log copies/ml = 2.10^6 IU/mL)
- high disease activity on liver biopsy
- HBV genotypes A and B

There are no consistent predictors of response for HBeAg-negative patients.

VI.1.5. On-treatment monitoring

During treatment with peg- IFN-α_{2a} blood count and liver panel should be monitored every 4 weeks. Dosage

of HBV DNA, HBeAg, HBeAb, HBsAg and HBsAb and TSH should be performed every 12 weeks.

VI.1.6. Tolerability and side effects

Peg- IFN-α_{2a} is reasonably well tolerated in HBeAg-positive and -negative patients and this tolerability is not modified by lamivudine co-administration (37,38,42). The most common side effects are flue-like symptoms (myalgia, fever, chills, headache and malaise), fatigue, anorexia, weight loss, emotional lability, hypo-and hyperthyroidism and hair loss. Severe myelodepression is uncommon (neutropenia < 1000/µL ; thrombocytopenia < 50,000/µL) except in patients who have diminished cell counts before starting the treatment. Depression occurs but with a lower frequency compared to patients with chronic hepatitis C (hepatitis B 5% versus 22% in hepatitis C patients) (47). Discontinuation of peg- IFN-α_{2a} because of side effects was necessary in $\leq$ 7% (37,38,42). Flares of ALT levels occur in about 30 to 40% during treatment. They represent a change in immunological response to HBV and are a predictor for favourable response (48). However, these flares can cause liver failure and decompensation, especially in patients with cirrhosis (7).

VI.1.7. Pregnancy

Because of antiproliferative effects is peg- IFN-α_{2a} contraindicated during pregnancy and the treatment should be stopped if the patient becomes pregnant (41).

VI.1.8. Costs of Peg- IFN-α_{2a} therapy

The cost for a 48 week treatment with Peg-IFN-α_{2a} is € 11,468 in Belgium. This is a finite treatment duration, while for nucleos(t)ide analogues indefinite treatment is sometimes necessary, which leads to higher costs than for Peg-IFN-α_{2a} (41).

Also the costs of disease activity and monitoring during and after treatment should be taken into consideration.

VI.2. *Nucleoside and nucleotide analogues in HBeAg-positive patients*

In Table 3 a summary of the efficacy of the different drugs is given for HBeAg-positive patients.

VI.2.1. Lamivudine (Zeffix®, Glaxo-SmithKline)

Lamivudine (3-thiacytidine) was the first L-nucleoside analogue licensed for use in chronic hepatitis B and has been considered the standard of therapy for this disease.

This drug has few and only minor side effects and the daily oral dose is 100 mg. The Belgian price for 1 month of therapy is € 62.40 (= € 749 per year).

Response to lamivudine

Loss of HBeAg was 32% vs. 11%, with HBeAg seroconversion observed in 16% to 18% after 1 year of treatment, compared with 4% to 6% of controls (49,50). HBeAg seroconversion rates increased with the duration of treatment to 50% after 5 years of continued treatment (51-54). Pretreatment serum ALT was the strongest predictor of response among HBeAg-positive patients. HBeAg seroconversion occurred in 2%, 9%, 21%, and 47% of patients with ALT levels within normal, 1-2 times normal, 2-5 times normal, and > 5 times normal range, respectively ; the corresponding seroconversion rates for patients in the placebo group were 0%, 5%, 11%, and 14%, respectively (55,56). HBV DNA was < 10^5 copies/mL after 1 year of treatment in 44% compared to 16% in placebo-treated patients. HBsAg loss was very rare (< 1% after 1 year). ALT normalization occurred in 41-72% of treated patients compared to 7-24% of controls. Finally, histology improved in 49% to 56% of treated patients and in 23% to 25% of controls. Lamivudine treatment for up to 6 years had an excellent safety profile in patients with HBeAg-positive compensated liver disease (54).

Durability of response to lamivudine

50%-77% of patients with HBeAg seroconversion had durable response. Several factors have been found to be associated with increased durability of lamivudine-induced HBeAg seroconversion, including longer duration of consolidation treatment (57,58).

For how long should Lamivudine be continued ?

The end point of treatment in HBeAg-positive patients is HBeAg seroconversion. Treatment can be discontinued in patients who have confirmed HBeAg seroconversion on 2 occasions and who received at least 6 months of consolidation therapy after the appearance of anti-HBe. In other patients, treatment should be continued if no resistance occurs (7,35).

Long-term outcome of Lamivudine-treated Patients

Because of the development of resistant strains, virological and biochemical response decreased with time (54). In patients in whom viral suppression could be maintained, necroinflammation and fibrosis were reduced and regression of cirrhosis was observed (59). Moreover, hepatic decompensation, liver-related mortality and development of HCC were lower in patients with sustained viral suppression (60).

VI.2.2. Adefovir dipivoxil (Hepsera®, UCB)

Adefovir dipivoxil is the pro-drug of adefovir, a nucleotide analogue. The 10 mg oral daily dose is well tolerated, even after 5 years of therapy (61). Higher daily doses ($\geq$ 30 mg) were associated with increased risk of renal damage and dose adjustment is recommended in patients with pre-treatment renal impairment (62). The Belgian price for 1 month of therapy is € 513.27 (= € 6,159 per year).

Response to adefovir

Patients treated for 48 weeks with adefovir 10 mg/d had a 12% chance of HBeAg seroconversion as com-

Table 3. — **Treatment results in HBeAg-positive patients with different medications**

In HBeAg+	Peg IFN	PegIFN + lami	Lamivudine	Adefovir	Entecavir	Telbivudine
HBV DNA decrease (log) EOT 6 m after stop	 - 4.5 - 2.4	 - 7.2 - 2.7	 - 5.8 - 1.9	 -3.5	 -6.9	 -6.5
HBV DNA < 400 IU/ml at EOT 6 m after stop	 25% 14%	 69% 14%	 40% 5%	 21% 8%	 67%	 60%
HBeAg loss EOT 6 m after stop	 30% 34%	 27% 28%	 22% 21%	 24%	 22%	 26%
HBeAg seroconversion EOT 6 m after stop 2 y 4 y	 27% 32%	 24% 27%	 16-20% 75%* 50%	 12% 90%*	 21% 70%*	 22% 34%
HBsAg loss 1 y 2 y 3 y 4 y			 20%**	 5%		
HBsAg seroconversion 1 y	3%	3%	0%	0%	0%	0%
ALT normalization EOT 6 m after stop	 39% 41%	 46% 39%	 40-75% 28%	 48%	 68%	 77%
Histological improvement 12 m	38%	41%	49-56%	53%	72%	65%
Resistance at 1 y 2 y 3 y 4 y 5 y	0% 0% 0% 0% 0%	4%	27% 42% 53% 70%	0% 2% 11% 18%	0% 0% 1%	4% 22%
Treatment duration	48 w	48 w	?	?	?	?
Cost for 1 y treatment	11,468 EUR		749 EUR	6,159 EUR	6,159 EUR°°	4,536 EUR°
Dosage	180 µg/w SC		100 mg/d po	10 mg/d po	0.5 mg/d po	600 mg/d po

* of those who had seroconversion.
** of those who had HBe seroconversion.
° : the prices given for telbivudine are those in neighbouring European countries, the price in Belgium has not yet been published.
°° : the price for entecavir 1 mg
Abbreviations : EOT : end of treatment ; SC : subcutaneous ; po : per os ; ALT : alanine aminostransferase ; lami : lamivudine ; pegIFN : pegylated interferon.

pared to 6% for the placebo group. Serum HBV DNA levels decreased by a mean of 3.5 $\log_{10}$ copies/mL (0.6 for placebo) with undetectable levels (< 400 copies/ mL) of serum HBV DNA in 21% vs. 0%. Normalization of ALT levels was observed in 16% and 48% of patients who received placebo or adefovir 10 mg/d, respectively. Finally, histological response was observed in 53% of patients who received adefovir 10 mg/d vs. 25% of those receiving placebo (63). HBe seroconversion increased after prolonged treatment to 33% and 46% after 96 and 144 weeks of treatment, respectively (35).

Decompensated cirrhosis

Adefovir has not been evaluated as a primary treatment for patients with decompensated cirrhosis (7).

For how long should adefovir be continued ?

As for lamivudine-treated patients, treatment with adefovir may be discontinued after confirmed HBeAg seroconversion and an additional 6 months of consolidation treatment. HBeAg seroconversion was maintained in approximately 92% of patients. Treatment may be continued in patients who have not achieved HBeAg

seroconversion but in whom HBV DNA levels remain suppressed (7).

Long-term outcome of adefovir-treated Patients

Long-term treatment was associated with a decrease in necroinflammation and fibrosis score in the vast majority of patients (61).

Adefovir Resistance

Resistance during adefovir treatment is lower as compared to lamivudine. In lamivudine-naïve patients, no adefovir-resistant mutations were reported after 1 year of treatment (63, 64). However, resistance emerged after prolonged treatment : 3%, 11% and 18% after respectively 2, 3 and 4 years of treatment (65). However, in patients with lamivudine-resistant HBV, adefovir resistance was approximately 20% after 1 year of adefovir monotherapy (66,67). In contrast, in lamivudine-resistant HBV patients treated with the combination of lamivudine and adefovir, there was no evidence of resistance to adefovir after 3 years (67).

VI.2.3. Entecavir (Baraclude® Bristol-Myers Squibb)

Entecavir is a deoxyguanosine (nucleoside) analogue with potent activity against the hepatitis B virus. The recommended oral daily dose is 0.5 mg for non-lamivudine-resistant patients and the profile is safe. Currently the 0.5 mg dose is not available in Belgium. The price of 1mg entecavir (the dose required for lamivudine resistant patients) is € 513.27 per month or € 6,159 per year.

Response

After 48 weeks of treatment, the rates of histological, virological, and biochemical improvement were significantly higher with entecavir than with lamivudine, with a similar safety profile. Histological improvement was observed in 72% in the entecavir group compared to 62% in the lamivudine group. The mean reduction in serum HBV DNA was greater with entecavir than with lamivudine (-6.9 vs. -5.4 $\log_{10}$ copies/mL). Undetectable serum HBV DNA levels (PCR assay) occurred in 67% vs. 36% and normalization of ALT levels was seen in 68% vs. 60%. HBeAg seroconversion was not significantly different in the two groups (21% vs. 18%) (68). After 2 years of treatment, HBeAg seroconversion was significantly higher in entecavir-treated patients (35).

Durability of Response

Among HBeAg-positive patients who underwent HBeAg seroconversion during the first year and who stopped treatment at week 48, approximately 70% of patients remained HBeAg-negative (7).

Entecavir Resistance

Virologic breakthrough was extremely rare in nucleos(t)ide-naive patients (< 1% of patients after 1 and 2 years of entecavir treatment, respectively) (68,69). Moreover, entecavir resistance was only observed in patients who harbored a lamivudine-resistant strain at entry. In patients previously treated with lamivudine who became refractory to lamivudine, resistance to entecavir was detected in 7% and in 16% of patients after 1 and 2 years of treatment (69,70). The cumulative probability of a virologic breakthrough due to entecavir resistance through 4 years was 0.8% in naïve and 39.5% in lamivudine refractory patients (71).

Predictors of Response

HBeAg seroconversion rates were lower in patients with normal ALT (12%) as compared to patients with mildly elevated ALT (23%) and patients with ALT > 5 times normal value (39%) (7).

VI.2.4. Telbivudine (Sebivo®, Novartis)

Telbivudine is a nucleoside analogue with potent antiviral activity against hepatitis B virus. The oral daily dose is 600 mg. Doses should be decreased in patients with renal failure (7). The safety profile of telbivudine is comparable to lamivudine. The price in other European countries is about € 378 for 1 month or € 4,536 per year. To date, the prices for Belgium have not been published yet.

Telbivudine Response

Patients treated for 1 year with telbivudine had a significantly greater mean reduction in HBV DNA levels (-6.01 vs -4.57 $\log_{10}$ copies/mL), clearance of detectable HBV DNA (61% vs. 32%) (Quantiplex branched DNA assay <3 Meq/mL), and normalization of ALT levels (86% vs. 63%) compared with lamivudine monotherapy (72). Also, after 2 years of treatment, the results were favourable for telbivudine. However, there was no difference in the rate of HBeAg loss at the end of 1 and 2 years of treatment : 26% vs. 23%, and 34% vs. 29% of patients who received telbivudine and lamivudine, respectively (73,74).

Telbivudine Resistance

Telbivudine is associated with a lower rate of drug resistance than lamivudine, but the resistance rate is substantial and increases after the first year of treatment. Genotypic resistance after 1 and 2 years of treatment was observed in 4.4% and 21.6% of HBeAg-positive patients, with viral breakthrough of 4.5% after 1 year (73,74).

VI.2.5. Combination therapy

Combination therapy has the theoretical advantage of higher efficacy and reduced occurrence of resistance. The major disadvantage is increased costs. At present there are no solid data indicating that combination therapy is superior to monotherapy in inducing sustained viral response (7). Furthermore, although resistance to lamivudine is reduced, it is not completely prevented. Currently, no data indicate that combination therapy reduces the risk of resistance to drugs with a low resistance rate. Therefore, at this time combination therapy is not advocated as first-line treatment in naïve HBeAg positive patients for treatment.

VI.3. *Nucleoside and nucleotide analogues in HBeAg negative patients*

Introduction

In HBeAg-negative patients, normalization of transaminases (biochemical response) and sustained HBV DNA suppression (virological response) and HBs seroconversion (in rare cases) are the only practical measures of response to therapy (35).

In Table 4 a summary of the efficacy of the different drugs for HBeAg-negative patients is given.

VI.3.1. Lamivudine (Zeffix®, Glaxo-SmithKline)

Biochemical and virological responses, even detected by sensitive PCR assays, ranged from 60 to 90% patients after 1 year of therapy (75-80), with histological improvement in the same proportion. Unfortunately, biochemical and virological relapses were observed in the majority of patients (around 90%) after stopping a 1 year course of therapy (76).

The association of lamivudine with pegylated interferon did not improve post-therapy response rate (42).

Table 4. — **Treatment results in HBeAg-negative patients with different medications**

In HBeAg-	Peg IFN	PegIFN + lami	Lamivudine	Adefovir	Entecavir	Telbivudine
HBV DNA decrease (log) EOT 6 m after stop	 - 4.1 - 2.3	 - 5 - 2.4	 - 4.2 - 1.6	 - 3.9	 - 5	 -5
HBV DNA < 400 IU/ml EOT 6 m after stop 2 y 5 y	 63% 19%	 87% 20%	 73% 7%	 51% (1yr) 71-79% 67%	 90%	 88-79% (1-2yr)
HBsAg loss 6 m after stop 5y	4%	3%	0%	 5%		0%
HBsAg Seroconversion 6 m after stop	 3%	 2%	 0%			
ALT normalization EOT 6 m after stop 2 y 5 y	 38% 59%	 49% 60%	 73% 44%	72% (1yr) 69%	 78%	 74-75% (1-2 yr)
Histological improvement 12 m 5 y	48%	38%	40%	64% 83-73%		
Resistance at 1 y 2 y 3 y 4 y 5 y	 0% 0% 0% 0% 0%	1%	18%	 5,9% 29%	 9%	 2,7% 9,8%
Durable undetectable DNA < 400 IU/ml 1 y 2 y 3 y	 15% 16% 18%	 12% 11% 13%				
Treatment duration	48 w	48w	?	?	?	?
Cost for 1 y treatment	11,468 EUR		749 EUR	6,159 EUR	6,159 EUR °°	4,536 EUR °
Dosage	180 µg/w SC		100 mg/d po	10 mg/d po	0.5 mg/d po	600 mg/d po

Abbreviations : EOT : end of treatment ; SC : subcutaneous ; po : per os ; ALT : alanine aminostransferase ; lami : lamivudine ; pegIFN : pegylated interferon

° : the prices given for telbivudine are those of our neighbouring European countries, the price in Belgium has yet not been published.

°° : the price for entecavir 1 mg and not for the 0.5 mg dose.

The same negative results were found for combination therapy with interferon α2b with lamivudine (81).

Around 30% of patients have a sustained biochemical and virological response after long term therapy up to 5 years (60,80,82,83). However, it seems that the majority of these patients relapses after discontinuation of lamivudine (75,84). The optimal duration of therapy and the outcome after discontinuation of lamivudine in patients with such prolonged remission is currently unknown (84,85).

Extending the duration of treatment was characterized by a progressive decrease of lamivudine efficacy and increasing rate of virological breakthroughs due to the appearance of YMDD mutant hepatitis B strains (75,77,78,82).

Resistance to lamivudine, characterized by a rise in HBV DNA, increased from 19-27% after 1 year of therapy (75,82,86) to 66% after 4 years (82). High pretreatment HBV DNA level was a strong predictive factor of drug resistant mutation (87). The emergence of lamivudine-resistant mutants can be associated with clinically significant hepatitis and worsening of liver histology (78,80), mainly in cirrhotic patients (60,82). To date there are no controlled data comparing the efficacy of starting with lamivudine plus salvage therapy upon lamivudine resistance against initial therapy with agents with a better resistance profile than lamivudine (85).

Because of the need for long treatment durations and high resistance profile, lamivudine is not an optimal first-line treatment in HBeAg-negative patients (7).

The efficacy of lamivudine did not differ in naïve or previously interferon-treated patients (60). Lamivudine retreatment in patients who developed YMDD mutants after a previous course of lamivudine is ineffective because of the rapid re-emergence of YMDD mutants (88).

VI.3.2. Adefovir dipivoxil (Hepsera®, UCB)

Efficacy

After 1 year of therapy, the efficacy of adefovir was significantly higher than placebo : normalisation of transaminases in 72% of patients treated with adefovir (versus 29% in the placebo group), undetectable serum HBV DNA by PCR assay in 51% (versus 0%) and improvement of liver histology in 64% (versus 33%) (89). However, the response is usually lost after discontinuation of such short therapy.

After 2 and 3 years of therapy, a decrease in serum HBV DNA of respectively 3.47 $\log_{10}$ and 3.63 $\log_{10}$ copies/mL was observed in patients treated with adefovir and HBV DNA levels were less than 1000 copies/mL in respectively 71% and 79%. Resistance mutations developed in 5.9% of patients after 3 years (90).

After 5 years of therapy, HBV DNA was less than 1000 copies/ml in 67% of patients, and transaminases were normal in 69%. Improvement of liver histology was observed with a decrease of inflammation and fibrosis in respectively 83% and 73% of patients. The cumulative probability of mutations was 29% ; the cumulative probability of mutations with virological resistance was 20% (61). Significant improvement of liver fibrosis, even with reversion of histologically proven cirrhosis, was observed after a 5 year period of therapy and was associated with HBs Ag loss in 5% of patients (91).

The main *advantage of adefovir* compared with lamivudine is the infrequent development of viral *resistance* (around 20% versus 66% after 4 years of therapy). Serum HBV DNA levels at 1 year seem to be a good predictive factor of development of resistance under long-term therapy with adefovir (92). The development of adefovir resistance is uncommon in the first 2 years of therapy but can be associated with biochemical and virological rebound and hepatic decompensation (93). As mentioned previously, in lamivudine-resistant HBV patients treated with the combination of lamivudine and adefovir, there was no evidence of resistance to adefovir after 3 years (67).

VI.3.3. Entecavir (Baraclude® Bristol-Myers Squibb)

Efficacy

After 2 years of therapy, it has been demonstrated that entecavir was more effective than lamivudine (94). Normalisation of transaminases was observed in 78% of patients treated with entecavir versus 71% of patients treated with lamivudine ; the decrease of serum DNA levels and improvement of liver histology were also significantly higher in patients treated with entecavir (respectively 90% versus 72% and 70% versus 61%). The safety profile was similar for the two drugs and no entecavir resistance was observed (94).

Resistance

Resistance to entecavir has been described mainly in patients with lamivudine resistance (69,71). Around 9% of lamivudine-resistant patients treated with entecavir developed resistance to entecavir after 2 year of therapy.

VI.3.4. Telbivudine (Sebivo®, Novartis)

Efficacy

A phase III study showed, after 1 year of therapy, a significantly higher percentage of patients with undetectable HBV DNA ($\leq$ 20,000 IU/ml) in patients treated with telbivudine compared to patients treated with lamivudine (88% versus 71%). Normalization of transaminases was similar in the two groups of patients (74% versus 79%) (72). After 2 years, patients treated with telbivudine had a significantly higher level of transaminases normalisation (75% versus 67%) and undetectable HBV DNA ($\leq$ 20,000 IU/ml) (79% versus 53%) in comparison to patients treated with lamivudine (95).

RESISTANCE

Telbivudine is associated with a lower rate of drug resistance than lamivudine. However, the resistance rate is substantial and increased exponentially after the first year of therapy. After 1 and 2 years of therapy, resistance was observed respectively in 2.7% and 8.6% in telbivudine treated patients compared to 9.8% and 21.9% in lamivudine-treated patients (72,95).
There are currently no long-term data available for telbivudine therapy beyond 2 years.

Recommendations for the treatment of HBeAg positive chronic hepatitis B (7).

– Patients with ALT > twice normal value or moderate/severe hepatitis on biopsy, and HBV DNA > 20,000 IU/mL should be considered for treatment (evidence level I).

- *Treatment should be delayed for 3 to 6 months in persons with compensated liver disease to determine if spontaneous HBeAg seroconversion occurs (evidence level II-2)*
- *Patients with icteric ALT flares should be promptly treated (evidence level III)*
- *Treatment may be initiated with any of the approved antiviral medications, but peg-IFN- α_{2a} and the nucleos(t)ide analogues with the highest efficacy in suppressing HBV DNA and lowest resistance rate (highest genetic barrier) are preferred as first-line option. Peg-IFN-α_{2a} should be considered as first-line in patients with high transaminases, low HBV DNA and active disease (evidence level I). Lamivudine is not considered a reasonable first-line treatment option because of the high risk of resistance with long-term therapy and proven inferiority to Peg-IFN-α_{2a} and entecavir in randomized clinical trials.*

– Patients with persistently normal or minimally elevated ALT (< twice normal value) should generally not be started on treatment (evidence level I)

– Liver biopsy may be considered in patients with fluctuating or minimally elevated ALT levels especially in those above 35-40 years of age. Treatment may be initiated if there is moderate or severe necroinflammation or significant fibrosis on liver biopsy (evidence level II-3).

Recommendations for treatment of HBeAg-negative chronic hepatitis B (7,35)

– Because HBeAg negative patients tend to have lower levels of serum HBV DNA than HBeAg positive patients but still may have active disease, it is recommended to treat patients who have HBV DNA levels of > 2,000 IU/mL and elevated transaminases (evidence level I)

– Peg-IFN-α_2, and the nucleos(t)ide analogues with the highest efficacy in suppressing HBV DNA and lowest resistance rate (highest genetic barrier) are preferred as first line options. Currently, nucleos(t)ide analogues treatment in HBeAg-negative patients should be viewed as indefinite and even lifelong. Lamivudine is not considered a reasonable first-line treatment option because of the high risk of resistance with long-term therapy and proven inferiority to PegIFN-α_{2a} and entecavir in randomized clinical trials.

– In patients with HBV DNA levels of > 2 000 IU/mL and normal transaminases, a liver biopsy has to be considered and the same therapeutic recommendations are proposed in case of histological active disease (evidence level I, II-2).

– In the absence of liver biopsy, follow-up of transaminases is recommended and therapy is proposed only in patients with elevated transaminases.

– Patients with HBV DNA levels ≤ 2 000 IU/mL and normal transaminases are considered as inactive HBs Ag carriers and no therapy is recommended (evidence level I).

VII. Management of antiviral resistance to current hepatitis B nucleos(t)ide analogue therapy

A major concern with long-term nucleo(t)side analogue (NA) treatment is the selection of antiviral-resistant mutations, marked by appearance of circulating HBV with reduced sensitivity to the antiviral agent (96-99). Among the approved NA therapies for hepatitis B, lamivudine is associated with the highest and entecavir with the lowest rate of drug resistance in NA-naïve patients. The rate at which resistant mutants are selected is related to pre-treatment serum HBV DNA level, rapidity of viral suppression, duration of treatment, and prior exposure to NA therapies (100). Table 5 summarizes the definition of terms commonly used in describing antiviral resistance (33).

Primary non response is defined as the inability of the NA treatment to reduce serum HBV DNA by > 1 $\log_{10}$ IU/mL after the first 6 months of NA administration (33).

Definitions of resistance

Resistance is typically categorized as genotypic, viral, and clinical. *Genotypic resistance* is based upon detection of HBV mutations that are associated with *in vitro* and *in vivo* resistance to antiviral agents (96-99). During treatment with nucleos(t)ide analogues, mutations in the polymerase gene of HBV can often be detected before there is a rise in HBV DNA or ALT levels (101,102). The difficulty with this definition is that it requires molecular testing which is expensive and may not be warranted clinically if there are no other signs of antiviral resistance. *Viral resistance or virological breakthrough* indicates that HBV DNA levels have increased, the usual criteria being ≥ 1 $\log_{10}$ IU/mL increase from a previous nadir on ≥ 2 occasions 1 month apart, in a patient who is compliant and still on treatment. *Clinical resistance or biochemical breakthrough* is defined by a rise in serum ALT levels. For patients in whom serum ALT levels fall into the normal

Table 5. — **Definition of Terms Relating to Antiviral Resistance/Non response to Nucleoside Analogue (NA) Treatment**

- Primary nonresponse : a ≤ 1 $\log_{10}$ drop in HBV DNA after 6 months
- Virologic breakthrough : increase in serum HBV DNA by ≥ 1 $\log_{10}$ (10-fold) above nadir after achieving virologic response, during continued treatment
- Biochemical breakthrough : increase in ALT above upper limit of normal after achieving normalization, during continued treatment
- Genotypic resistance : detection of mutations that have been shown in *in vitro* studies to confer resistance to the NA that is being administered
- Phenotypic resistance : in vitro confirmation that the mutation detected decreases susceptibility (as demonstrated by increase in inhibitory concentrations) to the NA administered

Abbreviations : NA : nucleosi(t)ide analogues ; ALT : alanine aminotransferase.

range during therapy, clinical resistance can be defined as a rise to above twice the upper limit of the normal range in conjunction with a rise in HBV DNA levels and/or genotypic resistance. These criteria become difficult to apply in the situation in which ALT levels never fall into the normal range, or were normal before therapy, or fluctuate spontaneously.

Virologic breakthrough is usually followed by *biochemical breakthrough*. Emergence of antiviral-resistant mutations can lead to negation of the initial response, and in some cases hepatitis flares and hepatic decompensation. Antiviral-resistant mutations can be detected months and sometimes years before biochemical breakthrough. Thus, early detection and intervention can prevent hepatitis flares and hepatic decompensation, and this is particularly important in patients who are immunocompromised and those with underlying cirrhosis. Waiting for *clinical breakthrough* should absolutely be abandoned.

Another potential consequence of antiviral-resistant mutations is cross-resistance with other NAs, thus limiting future treatment options.

Judicious use of NA in patients with chronic hepatitis B is the most effective prophylaxis against the development of antiviral-resistant HBV. Thus, patients with minimal disease and those who are unlikely to achieve sustained response should not be treated with NA, particularly if they are young (< 30 years). When possible, the most potent NA with the lowest rate of genotypic resistance should be administered and compliance reinforced.

Up to 30% of virologic breakthrough observed in clinical trials is related to medication noncompliance, thus, compliance should be ascertained before testing for genotypic resistance.

Location and terminology of antiviral resistant mutations (Table 6)

The pattern of development of HBV resistant mutants varies by chemical class of nucleos(t)ide analogues, which can be categorized as :

1. L-nucleosides, such as lamivudine, telbivudine, emtricitabine, and clevudine (no data on the management of resistance to the latter two are available).
2. Acyclic phosphonates such as adefovir and tenofovir : nucleotide analogues.
3. Cyclopente(a)nes such as entecavir : new nucleoside analogue.

Nomenclature in discussing HBV resistance uses an abbreviation for the gene region in lower case (rt for reverse transcriptase, c for HBcAg, s for HBsAg) followed by the wild-type amino acid symbol, its position in the gene region, and finally the mutant or variant amino acid symbol (7). Detection of resistant mutations usually requires sequencing of the polymerase gene, but various assays including reverse hybridization and restriction fragment length polymorphism have been developed that detect the more common mutations (103).

Table 6. — **Location and Terminology of Antiviral Resistant Mutations**

Agent	Domains within Polymerase region of HBV P gene and their Amino Acid Locations			
	A	B	C	D
Lamivudine and emtricitabine	rtL80V/I*	rtA181T rtV173L* rtL180M*	rtM204V/I/S	
Telbivudine			rtM204I	
Entecavir	rtI169T	rtT184S/A/I/L/F/G rtL180M**	rtS202G/I rtM204V/I**	rtM250V
Adefovir		rtA181V/T		rtN236T

* Secondary, compensatory mutation.
** Mutations selected during the first steps toward entecavir resistance.
Abbreviations : rt, reverse transcriptase.

VII.1. *Lamivudine Resistance*

Lamivudine (3-thiacytidine) was the first L-nucleoside analogue licensed for use in chronic hepatitis B and has been considered the standard of therapy for this disease. Unfortunately, lamivudine has a high rate of antiviral resistance (low genetic barrier), averaging 15% to 20% per year (98). For these reasons, long-term results of lamivudine therapy are poor.

The most common mutation involves substitution of methionine in the tyrosine-methionine-aspartate-aspartate (YMDD) motif of the HBV DNA polymerase for valine or isoleucine rtM204V/I, changing it to YVDD or YIDD (104,105). The rtM204V/I mutation is usually accompanied by a compensatory mutation upstream of the YMDD motif at rtL180M and/or rtV173L. The rtM204V/I mutations are considered primary resistant mutations that lower the susceptibility of HBV to lamivudine, while the rtL180M and rtV173L mutations are considered secondary or compensatory, allowing for the resistant mutant to replicate at a higher rate. Generally, development of the lamivudine resistant HBV makes other L-nucleosides ineffective.

Genotypic resistance can be detected in 14% to 32% after 1 year of lamivudine treatment (49,50,106) and increases with the duration of treatment to 60% to 70% after 5 years of treatment (51,54). Factors associated with an increased rate of lamivudine resistance include long duration of treatment, high pretreatment serum HBV DNA level, and a high level of residual virus after initiation of treatment (54,107). One study reported that the rate of lamivudine resistance was significantly higher in patients whose serum HBV DNA level exceeded 200 IU/ml (1,000 copies/ml) after 6 months of treatment compared to those with lower HBV DNA levels (63% vs. 13%) (107).

The clinical course of patients with lamivudine-resistant mutants is variable. *In vitro* studies showed that rtM204V/I mutation decreases replication fitness of HBV but compensatory mutations selected during continued treatment can restore replication fitness (108,109). Virologic breakthrough is usually followed by biochemical breakthrough and in some patients may be associated with acute exacerbations of liver disease and rarely hepatic decompensation and death (99,110,111). Exacerbations of hepatitis associated with the emergence of lamivudine-resistance have also been reported to be associated with HBeAg seroconversion, possibly via immune mediated mechanisms (111). Hepatitis flares may also occur after withdrawal of treatment due to rapid outgrowth of wild type virus, but two studies in Asia found that the occurrence of hepatitis flares and hepatic decompensation were similar among patients with lamivudine breakthrough who stopped or continued lamivudine treatment (112,113).

In patients who have breakthrough infection, testing for lamivudine-resistant mutants should be performed when possible. The vast majority of patients with confirmed lamivudine-resistance should receive rescue therapy with antiviral agents that are effective against lamivudine-resistant HBV mutants.

Patients in whom ALT and HBV DNA levels remain significantly lower than pretreatment values may be maintained on lamivudine temporarily without resorting to rescue therapy but it must be recognized that compensatory mutations will be selected during continued treatment leading to subsequent viral rebound and possibly hepatitis flares. Adefovir and tenofovir have a potent activity against lamivudine-resistant strains *in vitro* and *in vivo* (114,115) whereas entecavir has reduced efficacy against rtM204V/I mutants (116).

Adefovir for Lamivudine-resistant hepatitis B

a. *Decompensated cirrhosis and liver transplant recipients*

In a compassionate use study involving 128 patients with decompensated cirrhosis and 196 patients with recurrent hepatitis B after liver transplantation, addition of adefovir was associated with a 3-4 $\log_{10}$ reduction in serum HBV DNA levels, which was sustained throughout the course of treatment (117). Among the patients who completed 48 weeks of treatment, 81% of the pre- and 34% of the post-transplant patients had undetectable HBV DNA by PCR assay, and 76% and 49% respectively had normalization of ALT. The Child-Turcotte-Pugh score improved in more than 90% of the pre-transplant patients, and 1-year survival was 84% for the pre- and 93% for the post-transplant patients. Follow-up data on 226 pre-transplant patients showed that viral suppression was maintained in 65% of patients after 96 weeks of treatment with accompanying improvement in Child-Turcotte-Pugh scores as well as Model for End-stage Liver Disease (MELD) scores (118).

b. *Compensated liver disease* – While a pilot study in patients with compensated chronic hepatitis B and lamivudine resistance found no differences in HBV DNA suppression and ALT normalization in persons treated with the combination of lamivudine and adefovir compared to those receiving adefovir alone (119), patients who discontinued lamivudine were more likely to develop ALT flares during the first 12 weeks of adefovir monotherapy. In addition, recent data showed that switching to adefovir in patients with lamivudine-resistant HBV was associated with a higher risk of adefovir-resistance compared to adding-on adefovir (67,119-121). Thus, increasing evidence supports that *adding adefovir is better than switching to adefovir monotherapy* for patients with lamivudine-resistant HBV. For most patients with lamivudine-resistant mutants, particularly those with decompensated cirrhosis or recurrent hepatitis B post-transplant, long-term treatment will be required. Increasing data indicate that lamivudine should be continued indefinitely after the addition of adefovir to reduce the risk of adefovir resistance.

Tenofovir for Lamivudine-resistant hepatitis B

Tenofovir disoproxil fumarate is an acyclic adenine nucleotide with potent activity against both HBV and HIV *in vitro* and *in vivo.* Tenofovir appears to be more potent than adefovir and is effective against lamivudine-resistant strains of HBV DNA. Small comparative studies have been conducted in cohorts of patients with HBeAg-positive chronic hepatitis B and lamivudine-resistance without HIV co-infection. In a study with greater than 48 weeks of follow-up, all 35 patients (100%) treated with 300 mg of tenofovir daily were HBV DNA negative compared to only 44% (7 of 15 patients) treated with 10 mg of adefovir daily (122). Tenofovir could also rescue patients with lamivudine resistance who had an inadequate response to adefovir (123). Side effects and renal toxicity were comparable. These results suggest that tenofovir may be the agent of choice for lamivudine-resistant HBV and may ultimately replace adefovir in treatment of hepatitis B. Phase III trials of tenofovir for chronic hepatitis B are now under way.

Entecavir for Lamivudine-resistant hepatitis B

In a dose-finding phase II trial, entecavir was shown to be effective in suppressing lamivudine-resistant HBV but a higher dose of 1 mg was required (124). In a subsequent study, 286 HBeAg-positive patients with persistent viremia while on lamivudine were randomized to receive entecavir 1 mg or lamivudine 100 mg daily. At week 48, entecavir resulted in significantly higher rates of histologic (55% vs. 28%), virologic (21% vs. 1%) and biochemical (75% vs. 23%) responses compared to lamivudine (70). Lamivudine should be discontinued when patients are *switched to entecavir* to decrease the risk of entecavir resistance (see discussion on stepwise development of entecavir resistance).

Peginterferon for Lamivudine-resistant hepatitis B

Although previous exposure to lamivudine did not seem to affect the overall rates of HBeAg seroconversion of *Peg-IFN-*α_{2a} in HBeAg-positive patients in one study (38), *Peg-IFN-*α_{2b} therapy showed only marginal efficacy in patients harbouring lamivudine-induced YMDD lamivudine resistance (125). Analysis of the patient subgroup harbouring a YMDD-mutation should be included in all future studies of *Peg-IFN-*α in chronic hepatitis B to find out if *Peg-IFN-*α therapy is beneficial in this situation.

VII.2. *Telbivudine resistance*

Telbivudine (L-deoxythymidine) selects for mutations in the YMDD motif. To date, only rtM204I mutants without rtM204V/rtL180M M204I (but not M204V) have been observed (73). Although telbivudine is associated with a lower rate of drug resistance than lamivudine, the resistance rate is substantial and increases exponentially after the first year of treatment. In the phase III clinical trial, genotypic resistance after 1 and 2 years of treatment was observed in 4.4% and 21.6% of HBeAg-positive and in 2.7% and 8.6% of HBeAg-negative patients who received telbivudine compared to 9.1% and 35% of HBeAg-positive and 9.8% and 21.9% of HBeAg-negative patients who received lamivudine. The lower resistance rate in the lamivudine group compared to previously reported clinical trials on lamivudine (54) may be related to the fact that only patients with virologic breakthrough were tested and a less sensitive method (direct sequencing) was used for detection of resistant mutations. There is limited evidence from a small series that switching to or adding adefovir is a viable salvage option in telbivudine-treated patients exhibiting virological breakthrough (126).

VII.3. *Adefovir Resistance*

Adefovir dipivoxil was the second oral antiviral drug to be licensed for use in chronic hepatitis B. Resistance occurs at a slower rate during adefovir treatment compared to lamivudine and no adefovir-resistant mutations were found after 1 year of treatment in the patients who participated in the Phase III trials (64). However, novel mutations conferring resistance to adefovir have been described since (127,128). Aggregate data from 5 studies including 3 studies using the combination of lamivudine and adefovir in patients with lamivudine resistant HBV estimated the cumulative rate of adefovir resistance to be 15% by 192 weeks (92). The phase III trial in HBeAg-negative patients found that the cumulative probabilities of genotypic resistance to adefovir at 1, 2, 3, 4, and 5 years were 0, 3%, 11%, 18%, and 29%, respectively (129). Recent studies using more sensitive methods have reported detection of adefovir-resistant mutations after 1 year of treatment and rates of genotypic resistance exceeding 20% after 2 years of treatment (66,120). In these studies, adefovir resistance was predominantly found in patients with prior lamivudine resistance switched to adefovir monotherapy. The most common resistant mutations associated with adefovir therapy have been rtA181V/T and rtN236T, but several other single or multiple mutations have been described (120,130,131). *In vitro* studies showed that adefovir-resistant mutations decrease susceptibility 3-15 -fold only (127,128). Nevertheless, clinical studies found that viral rebound, hepatitis flares and even hepatic decompensation can occur (93). Risk factors for adefovir resistance that have been identified include suboptimal viral suppression and sequential monotherapy (66,120). Sequential treatment with lamivudine followed by adefovir had also been reported to select for dual-resistant HBV mutants (93). *In vitro* and clinical studies showed that rtN236T adefovir-resistant HBV mutants are susceptible to lamivudine and entecavir (128). The rtA181V/T mutation has reduced susceptibility to both lamivudine and entecavir in vitro, but remains sensitive to tenofovir (92). Indeed re-emergence

of lamivudine-resistant mutations has been reported soon after reintroduction of lamivudine in patients with prior lamivudine resistance and who developed adefovir resistance after being switched to adefovir monotherapy (93). There are anecdotal clinical cases where switching from adefovir to tenofovir resulted in a decrease in serum HBV DNA levels (123). This may be related to a higher dose of tenofovir being used 300 mg vs. adefovir 10 mg. One case series reported that two patients with adefovir-resistant HBV responded to entecavir with a decrease in serum HBV DNA to undetectable levels (120).

Adefovir and primary non-response. Some studies have reported that 20%-50% of patients receiving the 10 mg dose of adefovir have primary non-response indicating that the approved dose of adefovir may be suboptimal (120). Approximately 30% of patients who have no prior treatment with NAs have primary non-response to adefovir (132). Primary non-response correlates closely with high initial HBV DNA levels and may relate to an only moderate antiviral activity of adefovir. The presence of HBV variants with primary resistance to adefovir may also be important (133). Higher doses of adefovir have greater potency against HBV, but are associated with an unacceptably high rate of renal toxicity. Alternative treatments should be considered for patients who exhibit a primary non-response to adefovir.

VII.4. *Tenofovir resistance*

Tenofovir disoproxil fumarate has a potent activity against both HBV and HIV *in vitro* and *in vivo* (134). Tenofovir is licensed for use in HIV infection and has been evaluated extensively in patients with HIV/HBV co-infection (135-137). To date there are no data on the occurrence nor management of tenofovir resistance in HBV monoinfected patients.

VII.5. *Entecavir resistance*

Entecavir is an acyclic guanosine derivative with marked activity against HBV. In preliminary studies and in randomized controlled trials, entecavir showed excellent potency, high rates of suppression of HBV DNA levels and improvements in biochemical and histological features of disease (68,94). Rates of clearance of HBeAg and HBsAg at one year were similar to those of other nucleoside analogues. Virologic breakthrough was rare in nucleoside-naïve patients, and was observed in only 3% of patients by week 96 of entecavir treatment in the two phase III clinical trials. Resistant mutations to lamivudine and entecavir were detected in only two ($\leq$ 1%) patients while resistant mutations to lamivudine only were found in three patients (69). However, virologic breakthrough was detected in 7% of patients after 48 weeks, in 16% after 96 weeks and 38% after 3 years of treatment in the phase III trial of lamivudine-refractory patients (69,70,71). Resistance to entecavir appears to occur through a two-hit mechanism with initial selection of M204V/I mutation followed by amino acid substitutions at rtI169, rtT184, rtS202, or rtM250 (138,139). *In vitro* studies showed that the mutations at positions 169, 184, 202 or 250 on their own have minimal effect on susceptibility to entecavir, but susceptibility to entecavir is decreased 10-250-fold when one of these mutations is present with lamivudine resistant mutations, and $\geq$ 500-fold when two or more entecavir-resistant mutations are present with lamivudine-resistant mutations. Lamivudine should be discontinued when patients are switched to entecavir to decrease the risk of entecavir resistance. *In vitro* studies showed that entecavir-resistant mutations are susceptible to adefovir, but there are very few clinical data on the efficacy of adefovir in patients with entecavir-resistant HBV.

Recommendations for treating patients with HBV resistant mutants (Table 7)

1. *Avoid unnecessary treatment with nucleo(t)side analogues (NA) (evidence level III).*
2. *Initiate first-line treatment with a potent antiviral drug that has low rate of drug resistance (evidence level I, II-1)*
3. *Check for primary NA non response (after 6 months of treatment) and subsequently for NA viral resistance/breakthrough with 3 monthly PCR (evidence level III)*
4. *Always check for patient's compliance in case of primary non-response or viral resistance/breakthrough before changing to an alternative treatment regime (evidence level III).*
5. *In lamivudine-resistant patients adefovir add-on therapy should be preferred to adefovir sequential monotherapy. Alternatively, therapy can be switched to sequential entecavir monotherapy in case of contraindications to adefovir (evidence level II-3).*
6. *1st line adefovir resistant patients generally respond to add-on lamivudine therapy or switch to entecavir or telbivudine or tenofovir. Second-line adefovir use with resistance : discuss switch to entecavir or tenofovir (evidence level II-2, III).*
7. *Current data suggest that tenofovir is superior to adefovir in treatment of both naïve and lamivudine- or adefovir- resistant patients. Once this drug is licensed for chronic hep B monoinfection, tenofovir might replace adefovir (evidence level III).*

VIII. Follow up during and after treatment

VIII.1. *Follow-up during treatment*

VIII.1.1. Patients treated with *pegylated interferon* should have a monthly clinical and biochemical (transaminases, hemoglobin, leucocytes and polymorphonuclear, platelets) evaluation with a strict attention to compliance, to prevent and treat adverse effects. The virological monitoring (HBV DNA, HBe status and HBs

Table 7. — **Management of Nucleos(t)ide analogue resistance**

Lamivudine-resistance
• Add adefovir (preferred over switch to adefovir, especially in case of cirrhosis)
• Switch to entecavir (stop lamivudine because of risk of subsequent entecavir resistance)
• Potential future management : add/switch to tenofovir or switch to emtricitabine/tenofovir
Adefovir resistance
• Add lamivudine if no prior lamivudine resistance (preferred over switch to lamivudine)
• Switch to entecavir (if no prior lamivudine resistance)
• Potential future therapy : switch to emtricitabine / tenofovir
Entecavir resistance
• Add or switch to adefovir or tenofovir

status) and TSH is performed every 3 months. HBV DNA levels at weeks 12, 18, or 24 were not sufficiently predictive to develop a "stopping rule" similar to that developed in the treatment of chronic hepatitis C (140).

VIII.1.2. The following roadmap is proposed for the management of chronic hepatitis B patients treated with *nucleos(t)ide analogue* monotherapy.

– In patients starting treatment with nucleos(t)ide analogues, the first HBV DNA measurement (by a very sensitive method) should be done after *6 months*. This primary response depends on the compliance of the patient and/or on the efficacy of the drug used. If compliance is correct and if HBV replication is suppressed with $\geq 1 \log_{10}$ IU/mL or, preferably, to non-detectable values, monotherapy can be continued. Not achieving this early end point, in the presence of adequate compliance, should prompt a change of therapy : either switch to another more potent component, or add another component without cross-resistance to the first nucleos(t)ide analogue used. Patients are then monitored with HBV DNA measurements every 3 to 6 months in order to diagnose further response or non-response (141).

– Early detection of *viral drug resistance* ($\geq 1 \log_{10}$ IU/mL increase compared to the nadir on ≥ 2 occasions 1 month apart) has important clinical implications as discussed in the previous chapter. To detect emergence of viral resistance, HBV DNA level measurements by PCR should be done every 3 to 6 months depending on the treatment regimen (33).

– Patients, particularly cirrhotics and liver transplants, should have their renal function checked regularly because of the potential nephrotoxicity of some NA (especially adefovir). Dosage interval must be adjusted in case of renal failure for all NA shown in table 8.

VIII.2. *Follow-up after treatment*

VIII.2.1. Treatment is stopped after one year for *pegylated interferon* both in HBeAg-positive and negative patients. After the end of therapy, ALT and HBV markers (including HBV DNA) should be monitored to detect relapse and possibly HBsAg seroconversion.

VIII.2.2. For those on *nucleos(t)ide analogues*, treatment is stopped either after HBeAg-seroconversion, documented on two separate occasions (HBeAg-positive initially) or after HBsAg-seroconversion (HBeAg-negative initially) if an additional 6 months of consolidation treatment was given. In the latter category, treatment with NA might be stopped after a prolonged period of HBV DNA negativity but relapse is frequent.

– After stopping any therapy, relapses have to be detected by measuring liver enzymes after three months and HBV DNA after 6 months. Sustained virological response is defined as negative HBV DNA 6 months after stopping therapy.

– Most patients are being treated for many years with NA in order to control viral replication, prevent relapse after stopping therapy and lower the incidence of liver-related complications.

– Treated, non-treated and formerly treated patients are always at risk of developing hepatocellular carcinoma, those at higher risks being males over 50, those with a high level of DNA replication, ongoing hepatitis, cirrhotic patients, Africans > 20 y old, Asian women $\geq$ 50 y, Asian men $\geq$ 40 y.

– Reactivation of HBV is still possible, even after HBsAg seroconversion (142) in case of chemotherapy or immunosuppression. Therefore adequate monitoring is mandatory, tailored to the patient's profile (141).

– For non-responders, further monitoring is required in order to recognize delayed response or reactivation, to adapt or change the treatment and to prevent and detect complications at an early stage.

Recommendations for follow- up of chronic hepatitis B patients

1. *Initially, patients with chronic hepatitis B must have a thorough clinical, biochemical and virological evaluation and counselling in order to classify them according to the four forms of HBs Ag carriers either HBeAg-positive (immunotolerant or chronic hepatitis due to wild type virus) and HBeA-negative (inactive carrier or chronic hepatitis due to the core/pre-core-mutants) (evidence level III).*
2. *Well-characterized immune tolerants and inactive carriers may neither be submitted to liver biopsy nor to treatment but clinical, biochemical and virological follow-up is necessary together with screening for hepatocellular carcinoma (evidence level III).*
3. *HBeAg-positive or-negative patients with chronic ALT elevation and HBV DNA above 10^{4-5} copies/ml (2,000-20,000 IU/mL) should be assessed after liver biopsy and treatment should be considered in the presence of necroinflammation and/or fibrosis (evidence level I). In cirrhotic patients, any HBV DNA elevation is a criterion for considering treatment.*
4. *During therapy, clinical symptoms, ALT, HBeAg, HBV DNA should be monitored regularly. Renal function should be monitored especially if adefovir is*

Table 8. — **Pegylated interferon alfa 2a and Nucleos(t)ide° dose-adaptation in case of impaired renal function**

Drug	Standard dose	Creatinine clearance mL/min and dose adaptation			
Pegylated IFN alfa 2a		< 40 or dialysis			
	180 µg/week SC	135 µg/week	SC		
Lamivudine		30-50	15-30	5-15	< 5
	100 mg/d	50 mg/d	25 mg/d	15 mg/d	10 mg/d
Adefovir		20-49	10-19	Dialysis	
	10 mg/d	10 mg every 48 h	10 mg every 72 h	10 mg every 7 d after dialysis	
Entecavir		20-49	10-19	Dialysis or CAPD	
Naïve pts	0.5 mg/d	0.25 mg/d	0.15 mg/d	0.05 mg/d after dialysis	
Lamivudine resistant pts	1 mg/d	0.5 mg/d	0.3 mg/d	0.1 mg/d after dialysis	
Telbivudine*		30-49	< 30	Dialysis	
	600 mg/d	600 mg every 48 h	600 mg every 72 h	600 mg every 96 h after dialysis	
Tenofovir disoproxil		30-49	10-29	Dialysis	
	245 mg/d	245 mg/d every 48 h	245 mg every 7 d after	245 mg every 72-96 h	

* Telbivudine dose adaptation is based on extrapolation and is possibly suboptimal and is not clinically evaluated. The clinical response and renal function should be checked regularly.
° Nucleos(t)ide analogues are given orally.
Abbreviations : pts : patients ; CAPD : continuous ambulatory peritoneal dialysis ; SC : subcutaneous.

used. During pegylated interferon, monitoring of the adverse effects is mandatory (evidence level III).

5. *Early monitoring of the viral load is mandatory in order to identify primary non-response and to prevent/detect viral resistance to NA. Monitoring of HBV DNA is done after 6 months to assess primary non-response (< 1 log_{10} IU/mL decrease in HBV DNA) and then every 3-6 months (depending the drug used). The therapy is adapted accordingly (evidence level III).*
6. *Criteria for stopping therapy include repeated (at least on 2 subsequent assessments with 6 months interval) documentation of HBe or HBs seroconversion and HBV DNA negativity (evidence level I).*
7. *Sustained virological response must be assessed 6 months after stopping therapy (evidence level I). Virological relapse is not infrequent and should lead to reinstitution of an appropriate therapy (evidence level III).*
8. *All non-responders and/or non-treated patients with persistence of HBsAg and HBV DNA positivity should have adequate follow-up in order to prevent and detect early complications and HCC surveillance every 6 months (evidence level II-2).*

IX. Special groups : Hepatitis B co-infected patients

IX.1. *HBV/HCV coinfected patients*

Epidemiological aspects

As hepatitis B and C share modes of transmission, their combined occurrence is not uncommon, particularly in areas where the two viruses are endemic and in subjects with high risk of parenteral infection e.g. injection drug users (143,144). Seroprevalence studies have shown that approximately 10-20% of patients with chronic hepatitis B are also coinfected by HCV, whereas 2-10% of anti-HCV-positive patients are reported to have markers of HBV infection (145).

Natural history of coinfection with HBV and HCV

Dual infection tends to aggravate the severity and progression of chronic liver disease (144) and presents a much higher relative risk for the development of hepatocellular carcinoma (146).

Interaction of HBV and HCV

Several studies have shown that HBV and HCV interact with each other and affect immune responses. In vitro studies indicate that HCV is capable of suppressing HBV activity and that this inhibitory effect is mediated by the HCV core protein (147). *In vivo* studies also indicate a possible interplay between the two viruses : some cases confirm a prevalent role of HCV (148), while other reports suggest a reciprocal interference or dominant effect of HBV (144). This confusing information about HBV and HCV behaviour in case of dual infection has been generated primarily by cross-sectional studies. In a recent Italian multicenter study that longitudinally examined 133 HBsAg and anti-HCV positive patients for 1 year showed that a wide and complex spectrum of virological profiles may occur in cases of coinfection (149). In fact, about one third of the cases presented alternate phases of inhibition and recurrence of the activity of one or both viruses, as revealed by broad changes over time of the amount of circulating HBV DNA or, less frequently, HCV RNA. Moreover, the

longitudinal profile of each virus in all the cases with fluctuating virological patterns appeared to be totally independent of the viraemia levels of the other virus. It was concluded that a correct diagnosis should be performed by serially repeating the virological tests over one year.

Treatment of HBV/HCV coinfection

As anti-HCV-positive individuals are excluded from studies on HBV-related liver disease, very little information is currently available on the therapeutic approach to the HBV/HCV-coinfected population. The studies performed in this field agree in considering these chronic hepatitis patients particularly difficult to cure. Considering that HBV/HCV coinfection is highly prevalent in many areas of the world, and in view of the severity of the associated liver diseases, it appears particularly urgent to design and perform therapeutic trials for the definition of the best approaches for treatment of these patients.

There is no currently established standard of care for HBV/HCV coinfected patients. In general, the same treatment criteria should be applied to patients with coinfection as are applied to monoinfected patients. As with HBV and HCV, initiation of therapy is recommended in case of active chronic hepatitis or cirrhosis prior to decompensation.

At present, the therapeutic schedule should be tailored to each individual patient on the basis of his/her own virological profile. On the basis of the study of Raimondo *et al.* (149), both HBV and HCV viraemia levels should be longitudinally evaluated before deciding the most appropriate treatment of coinfected patients (150).

Patients should be assessed to determine which virus appears to be dominant, based on serologic markers and levels of viraemia, and then treated accordingly (evidence level III).

- Hence, patients with HBV DNA > 20,000 IU/mL or higher and undetectable HCV RNA should be treated for HBV.
- A recent study showed that HBV/HCV coinfected patients with predominant HCV infection respond as well as patients with chronic HCV monoinfection to interferon-alpha plus ribavirin therapy (151). It seems therefore appropriate to treat these patients according to the current standards for therapy (pegylated interferon alpha + ribavirin) of chronic hepatitis C (152).
- HBV/HCV coinfected patients with detectable HCV RNA and high levels of HBV DNA should start with pegylated interferon-alpha and ribavirin in standard doses. In the absence of any effect on HBV DNA after 1 year, a nucleos(t)ide analogue might be started. If there is a contraindication for Peg-IFN it is recommended to use as first-line therapy the most potent available compound in suppressing HBV DNA associated with the lowest incidence of resistance (7,35).
- Once therapy is started, the virological follow-up must be continued in order to recognize possible reactivation of a previously quiescent infection, as has been reported sporadically, and to adapt the therapy to the new virological scenario.

Recommendations for HBV/HCV coinfected patients (evidence level III) :

- *Patients with HBV DNA of 20,000 IU/mL or higher and undetectable HCV RNA should be treated for HBV.*
- *HBV/HCV coinfected patients with predominant HCV infection : treat these patients according to the current standards for therapy (pegylated interferon + ribavirin) of chronic hepatitis C* (152).
- *HBV/HCV coinfected patients with detectable HCV RNA and high levels of HBV DNA should start with Peg-IFN-alpha and ribavirin in standard doses. In the absence of any effect on HBV DNA after 1 year, a nucleos(t)ide analogue may be started. If there is a contraindication for Peg-IFN it is recommended to use as first-line therapy the most potent available compound for suppressing HBV DNA associated with the lowest incidence of resistance* (7,35).
- *Once therapy is started, the virological follow-up must be continued in order to recognize possible reactivation of a previously quiescent infection and to adapt the therapy to the new virological scenario.*

IX.2. *HBV/HDV coinfected patients*

Epidemiological aspects

Hepatitis Delta virus (HDV) is a sub-viral agent that is dependent for its life cycle on the hepatitis B virus. The help it obtains from HBV is limited to the sharing of envelope proteins. It is speculated that worldwide around 15-20 million people (about 5% of the HBV carriers) are coinfected with HBV/HDV (153). HDV is spread in the same ways as HBV through materno-neonatal, parenteral or sexual exposure to blood or body fluids. Endemic zones are the Amazone basin, Eastern Europe, Asia and the Mediterranean area. In the latter area, implementation of HBV vaccination is contributing to the current decline of HDV (154). In Italy, for example, the prevalence of anti-HDV chronic HBsAg carriers with chronic liver disease has declined from 25% in 1985 (155) to less than 10% in recent years (154). However, in southern Europe there is still residual manifestation of the disease in patients infected years ago when HDV was endemic.

Furthermore in our neighbouring countries Germany and France, it is not a disappearing disease, mainly on account of immigration of people from endemic areas. In these countries, surveillance for HDV is being carried out (156, 157).

Natural history of HBV/HDV coinfection

HDV infection can cause severe acute and chronic liver disease with a chronicity rate reaching 70-90% in

case of superinfection (the most prevalent form of infection) and a common progression to cirrhosis (60-70%) accounting for frequent evolution to end-stage liver disease and hepatocellular carcinoma (158).

INTERACTION OF HBV AND HDV

Usually, HBV replication is inhibited by HDV and HBV DNA can be detected in serum of most HDV coinfected patients only by sensitive assays. Patients with chronic HDV and high levels of HBV DNA in blood were shown to have more severe disease than those with low viraemia (159).

TREATMENT OF HDV

No specific inhibitor of HDV has so far been developed. Drugs that specifically block HBV have little or no effect on HDV replication. This is most likely due to the fact that the only helper function HBV provides to HDV is the HBsAg envelope which is efficiently expressed in most HBV carriers regardless of the level of HBV replication. Thus, an effective anti-HDV therapy would require a marked suppression of HBsAg, but current therapies for HBV do not achieve this. Lamivudine, a nucleoside analogue that potently inhibits HBV replication, has no effect on HDV viraemia or liver diease activity in patients with chronic HDV (160).

The only treatment for chronic HDV is interferon-alpha, currently the only licensed drug for this disease. However, the response is limited and variable. Up to 70% of patients may reach normal transaminase levels while on therapy, but the relapse rate is high after discontinuation. Therapeutic efficacy increases when higher doses (9 MU thrice weekly tiw) are administered for prolonged periods (12-24 months) (154,155). A one-year course of interferon-alpha is associated with only 10-20% chance of sustained HDV clearance (161) and 10% chance of HBV elimination (162). Unfortunately, compliance with an intensive interferon therapy has been poor because of the dose-dependent development of side effects. Recently, however, it was shown that interferon-alpha significantly improves long-term clinical outcome and survival of patients with chronic HDV (163).

Two recent trials support the use of peg-IFN-α_{2b} (1.5 µg/kg/week) in chronic HDV in 14 patients treated for 12 months (164) and 38 patients treated for 72 weeks (165). In the former study, virological response was 8/14 (57%) at the end of treatment, and sustained virological response 6 months after treatment was 6/14 (43%). In the latter study, virological response was obtained in 5/38 (13%) ; in 8 other patients (21%) HDV viraemia diminished to less than 1000 copies/mL. At the end of follow-up (6 months after treatment), 8 patients (21%) were HDV RNA negative. Adding ribavirin had no effect on the viral clearance rate.

Recommendations for HDV/HBV treatment (evidence level II-3)

- *Based on the available data, high dose interferon or pegylated interferon for 1 year appears to have a long-term beneficial effect in patients with chronic HDV* (7).
- *In HBV/HDV coinfected patients with active HBV replication, where interferon-based therapy is contraindicated or who are not responsive to this treatment, therapy with a nucleoside/nucleotide seems appropriate. For this purpose, it is recommended to use as first-line therapy the most potent available compound for suppressing HBV DNA associated with the lowest incidence of resistance.*
- *For end-stage HDV liver disease, liver transplantation is a valid option ; the risk of re-infection is lower for HDV than for HBV under long-term administration of hyperimmune serum (HBIG) against HBsAg* (166).

IX.3. *HBV/HIV co-infected patients*

GENERAL CONSIDERATIONS

Between 6% and 13% of patients infected with HIV are also co-infected with HBV (7). Patients with HIV infection develop chronic HBV infection more frequently and, in addition, the natural history of chronic HBV infection is modified in these patients (167). In particular, individuals with HIV and HBV have more severe liver disease with increased risk of cirrhosis and end-stage liver disease as well as a higher incidence of liver-related mortality (7,167-170). Furthermore, HIV/HBV co-infected patients tend to have higher HBV DNA levels and lower serum aminotransferase levels making them less useful in assessing the need for therapy (168). One has to keep in mind that a substantial proportion (10 to 45%) of HIV-infected patients with isolated anti-HBc antibodies have detectable levels of HBV-DNA and necroinflammation on liver biopsy (171-174). Therefore, it is recommended to test all HIV-infected patients with positive HBsAg and/or anti-HBc serology for HBV DNA in order to detect so called "occult" chronic HBV-infection.

TREATMENT

The treatment end points for HBV infection in the HIV population are suppression of viral replication and improvement of liver disease. However, immune control, i.e. seroconversion to anti-HBe and anti-HBs, is a rare event in HIV-infected patients. Long-term therapy is therefore the rule (168). Liver biopsy should be considered before making decisions whether to treat or not, given the limited value of serum aminotransferase levels in assessing disease activity and a non-negligible proportion of patients having occult HBV infection. It seems reasonable to base HBV treatment decisions on whether or not HIV treatment is required or planned in the near future. Most of the recommendations are based on expert opinion since only a few studies, most of them non-randomized with only small numbers of patients included, have focused on the HIV/HBV co-infected population.

HIV/HBV co-infected patients not requiring HIV treatment

Treatment guidelines for HBV infection in this group of patients can be based on standard criteria. However, these patients should not receive HBV medications with activity against HIV in order to avoid selecting resistant HIV viruses. Telbivudine, a molecule that does not target HIV, should also not be used in HBV/HIV co-infected patients because of the risk of selection of M204I mutation in the YMDD motif that confers cross-resistance with lamivudine, often used in combination regimens in patients on HAART (7,168).

Studies conducted with standard interferon alpha treatment reported lower response rates in the co-infected population compared to HBV monoinfection (7,167). Although it is expected that pegylated interferon alpha will have better efficacy than standard interferon alpha, it has not been tested in clinical trials in HIV/HBV co-infected patients. As a general rule, pegylated interferon alpha may be considered as a first-line option in HBeAg-positive patients with CD4 cell counts above 500 cells/μL given the limited duration of treatment (7). Nevertheless, one has to bear in mind that this drug is most successful in patients with high alanine aminotransferase levels and low HBV viral loads, both of which are uncommon in HIV infection. As an alternative, adefovir can also be used in this setting.

A recent paper by Mc Mahon reports that entecavir inhibits HIV-1 replication and may select for virus mutations (M184V in HIV-1 reverse transcriptase) which may lead to emtricitabine and lamivudine resistance (175,176).

Adefovir is the first-line option in HBeAg-positive patients with low CD4 cell counts or who are HBeAg-negative regardless of the CD4 cell counts (7). Entecavir is no longer recommended in the situation where HBV treatment is necessary without HIV treatment (175,176).

HIV/HBV co-infected patients requiring HIV treatment

For patients in whom HAART is indicated, it is best to choose a regimen that includes drugs with activity against HBV. Drugs with activity against both HIV and HBV include lamivudine, emtricitabine and tenofovir (7). However, patients who are already on HAART which does not include a drug with anti-HBV activity may be treated with pegylated interferon alpha, adefovir or entecavir (7). The rate of HBV resistance to lamivudine used as the sole agent with anti-HBV activity is unacceptably high (90% at 4 years) and should be avoided (171). Only 1 patient with resistance to tenofovir has been reported so far (177) whereas no data on resistance to emtricitabine in HIV/HBV co-infected patients are available to date. As described above, entecavir may inhibit HIV-1 replication and may lead to the development of resistance (175,176). Most experts recommend using a combination of two drugs with anti-HBV activity such as tenofovir plus lamivudine or emtricitabine (178,179). For combination therapy, including tenofovir appears to reduce the rate of lamivudine resistance (180). In patients who are already on HAART and in whom a lamivudine resistant virus has emerged, tenofovir or alternatively adefovir should be added (7,168, 178,181). Although the resistance rates to adefovir increase over time (18% at 4 years) in patients on monotherapy (127), this trend has not been confirmed in HIV/HBV co-infected patients receiving both molecules despite the presence of YMDD mutations at baseline (178). Finally, when HAART regimens are changed, it is important not to discontinue drugs that are effective against HBV without substituting another drug with anti-HBV activity.

Recommendations for HBV/HIV coïnfected patients (7)
HIV/HBV co-infected patients not requiring HIV treatment (evidence level II-3)

- *Pegylated interferon alpha may be considered as a first-line option in HBeAg-positive patients with CD4 cell counts above 500 cells/μL given the limited duration of treatment. Nevertheless, one has to bear in mind that this drug is most successful in patients with high ALT levels and low HBV viral load, both of which are uncommon in HIV infection. As an alternative, adefovir can also be used in this setting.*
- *Adefovir is the first-line option in HBeAg-positive patients with low CD4 cell counts or who are HBeAg-negative regardless of the CD4 cell counts or in patients with contraindications to pegylated interferon alpha.*

HIV/HBV co-infected patients requiring HIV treatment (evidence level II-3)

- *For patients in whom HAART is indicated, a regimen that includes drugs with activity against both HIV and HBV are preferred : lamivudine + tenofovir or emtricitabine + tenofovir.*
- *Patients on HAART without an anti-HBV activity drug may be treated with pegylated interferon alpha, adefovir or entecavir.*
- *In patients who are on HAART and in whom a lamivudine resistant virus has emerged, tenofovir or alternatively adefovir should be added.*
- *If HAART regimens are changed, it is important not to discontinue drugs that are effective against HBV without substituting another drug with anti-HBV activity.*
- *Anti-HBV treatment should be continued until the patients has achieved anti-HBe or anti-HBs seroconversion and has completed his consolidation treatment of 6 months.*

X. Special HBV patient populations

X.1. *Management of acute hepatitis B*

There is currently no recommendation to treat acute hepatitis B. In adults, most acute HBV infections will be

self-limiting and do not need antiviral treatment. Nevertheless, in some small series of severe acute hepatitis B, early therapy with nucleoside analogues seemed to be of some benefit (182-184). Given the serious risk of severe acute hepatitis B and the good safety profile of nucleoside analogues, antiviral treatment should be offered at the first sign of severe injury or liver failure (decrease of prothrombin (PT) activity and/or encephalopathy) (41). Nevertheless, it is important to point out that in a recent study comparing lamivudine versus placebo for acute HBV, there was no major clinical or biochemical improvement in the lamivudine arm (185).

Recommendations for acute hepatitis B (7,35)

- *Typical acute hepatitis B does not need antiviral therapy (evidence level III).*
- *Patients with severe acute hepatitis B (PT activity under 50% and/or hepatic encephalopathy) can be treated by nucleot(s)ide analogues (lamivudine, telbivudine or entecavir are preferred) as soon as the first sign of liver failure occurs. In these cases the patient should also be considered for liver transplantation (evidence level III)*
- *Patients treated with nucleos(t)ide analogue for their acute HBV, should maintain their treatment until HBsAg clearance is observed and confirmed. If transplanted, therapy should be maintained at least 1 year + hepatitis B immune globulins (HBIG) (see below) (evidence level II-1).*

X.2. *Management of compensated and decompensated HBV cirrhosis*

Standard Interferon-α and Pegylated-Interferon-α

Approximately 20-40% of patients with chronic hepatitis B will develop a flare of ALT levels during treatment by standard Interferon-α. In patients with cirrhosis, this flare may precipitate hepatic decompensation (7,35). In *compensated cirrhosis* (with HBe Ag+), the use of Interferon-α seems to be safe and produces similar results to those in non-cirrhotic patients (7,35). In *decompensated cirrhosis* (with HBe Ag+), two studies reported minimal benefit, but significant side effects due to bacterial infection and exacerbation of liver disease (186,187).

There are limited data documenting Peg-Interferon-α safety and efficacy in advanced liver disease due to HBV infection (7,35,41).

Lamivudine

Lamivudine has demonstrated its safety and efficacy in *compensated HBV cirrhosis*. However, these patients require prolonged treatment and the long-term benefit of lamivudine treatment is impaired by rapid emergence of resistant HBV mutants. In a large randomized controlled trial including 651 Asian patients (HBe Ag+ or HBV DNA > 140,000 IU/mL) with advanced liver disease, lamivudine treatment significantly slowed the disease progression and decreased the HCC emergence (25). Nevertheless, the benefit was only evident in half of the patients who did not develop viral resistance (25).

In *decompensated cirrhosis*, lamivudine was well tolerated and could stabilize or improve liver function thereby obviating or delaying the need for liver transplantation (188,189). However, it should be remembered that it takes several months (3-6 m) before clinical improvement occurs (7). Furthermore, HCC can occur even among patients with clinical improvement and therefore, continued HCC surveillance is necessary.

Adefovir

Adefovir has not been evaluated as a primary treatment for patients with cirrhosis. However, in patients with *decompensated cirrhosis* and resistant to lamivudine, the addition of adefovir to lamivudine was associated with a 3-4 $\log_{10}$ decrease of the viral load which became undetectable in 81 % of patients who finished a 48 week treatment (117). Moreover, in pre-transplant patients, viral suppression was maintained in 65% of these patients after 96 weeks of treatment (118).

Recent data have shown that switching to adefovir in patients resistant to lamivudine was associated with higher risk of adefovir resistance compared to adding adefovir to lamivudine (93). Increasing data also indicate that lamivudine should be continued indefinitely after the addition of adefovir to reduce the risk of adefovir resistance (7). Possible adefovir nephrotoxicity has been reported in up to 28% of patients with *decompensated cirrhosis* and therefore, frequent monitoring of serum creatinine is necessary (117).

Entecavir

Entecavir has more potent and more rapid antiviral effects than lamivudine and adefovir and at the dose of 1 mg/day entecavir is active against lamivudine-resistant mutants. This nucleoside analogue is promising for HBV *decompensated cirrhosis* and studies on safety and efficacy are ongoing.

Recommendations for compensated cirrhosis (7,35)

- *Patients with compensated cirrhosis should be considered for treatment if HBV DNA is > 2000 IU/mL regardless of ALT levels. Given the risk of Interferon-induced flares, nucleo(t)side analogues should be preferred. In view of the need for long term therapy and because of the rapid emergence of resistant mutants with lamivudine, first-line treatment with adefovir or entecavir should be started (evidence level II-2, II-3).*

Recommendations for decompensated cirrhosis (7,35)

- *Interferon-α and Peg-Interferon-α should not be used in patients with decompensated cirrhosis (evidence level II-3).*
- *In decompensated cirrhosis, independent of the viral load, antiviral treatment should be promptly initiated*

with nucleos(t)ide analogues producing rapid viral suppression and low risk of resistance. Currently, the combination of lamivudine and adefovir should be recommended for achieving a rapid effect and reducing the emergence of resistance. Entecavir is a promising treatment in this setting but clinical data are currently lacking (evidence level II-1, II-2, III).

- *Evaluation for liver transplantation should be considered (evidence level III)*

X.3. *Hepatitis B virus reactivation during immune suppressive treatment*

There are currently no accepted guidelines for the screening and prevention of HBV reactivation in patients undergoing chemo- or immunotherapy. As a result, a large number of patients with overt but also occult HBV infection are exposed to an unnecessary risk.

Definition

In recent guidelines, HBV reactivation has been defined as : "The reappearance of active necroinflammatory disease of the liver in a person known to have the inactive HBsAg carrier state or resolved hepatitis B" (7,35). A more exact definition could be : "The relapse or enhancement of viral replication during or immediately after immunosuppressive therapy resulting in liver damage" (190,191).

Magnitude of the problem

HBV reactivation has been mainly reported in patients with detectable HBsAg. However, patients negative for HBsAg but positive for anti-HBc alone or with anti-HBs have also, though less frequently, been reported to reactivate HBV infection (192).

The condition ranges from asymptomatic elevation of HBV DNA levels to fatal hepatitis. In case of icteric hepatitis, fatality rates ranged between 5% and 40% (191). Hepatitis is not the only threat of HBV reactivation. Indeed, in a large proportion of cases, anticancer treatment was interrupted due to hepatitis and the patient prognosis was jeopardized.

The magnitude of the problem depends upon the degree of HBV endemicity. HBV reactivation is a major problem in endemic countries (East Asia and Africa) where the prevalence of HBs Ag carriers ranges from 8-25% and the prevalence of anti-HBc is around 40% (190). In Belgium, where the prevalence of HBs Ag carriers is around 0.5% and the prevalence of patients with previous infection is around 3%, the incidence is less impressive. It must be taken into account that patients with haematological malignancy or with chronic rheumatic disease may have a higher risk of previous exposure to HBV. Therefore, in our country, it may be anticipated that up to 5% of patients treated by immunosuppressive and chemotherapeutic agents are at risk of HBV reactivation.

Diagnosis

In an appropriate clinical setting, and after exclusion of other possible causes, the diagnosis will be recognized as (190,191).

1) "Hepatitis" : a sudden rise of ALT over 5-fold the upper limit of normal or over 3-fold the baseline level.
2) "HBV reactivation" : a 10-fold rise in viral load. It is crucial to point out that the rise of HBV DNA precedes the ALT elevation and that HBV DNA levels tend to decrease when ALT levels increase. As a consequence, HBV DNA may be undetectable at the time of ALT elevation (190,191).

HBV reactivation most frequently follows cessation of chemotherapy, but may occur during chemotherapy. The reported interval ranged from 4 to 36 weeks (median 16 weeks) from initiation of chemotherapy (190).

Patients at risk

Patients with overt *HBV* infection (HBs Ag positive), patients with occult infection (HBs Ag negative, but serum and/or tissues HBV DNA positive) and even patients with resolved infection (anti-HBs positive, anti-HBc positive and HBV DNA negative) are at risk of HBV reactivation when receiving chemo- or immunotherapy. In patients with overt or occult infection, the risk of reactivation is estimated around 25% and depends upon the intensity of the immune depressive treatment (190-192). Of course, persons who are HBeAg positive or have pre-chemotherapy HBV viral load $\geq$ 2000 IU/mL are at higher risk of developing reactivation (193).

Also *young* persons and the *male gender* are more at risk (190,191,194). Besides the HBV status, gender and age, the *underlying disease* for which immunosuppressive or chemotherapy should be given and the *type of drug* are important to determine the risk of reactivation.

The underlying disease and drug type as risk factor :

1) Patients with lymphoma, leukaemia and other hematological malignancies. Lymphoma is the commonest cancer associated with HBV reactivation. In this group, raised transaminases will occur in more than 50 % of inactive HBs Ag carriers, jaundice in 10 % and liver related death in 5 % (191).
2) Patients receiving haematopoietic stem cell transplantation : in this group, HBV reactivation has been reported in over 50 % of patients (191).
3) Patients treated for solid cancers such as breast cancer, gastric cancer, renal cancer : the importance of the risk is less known.
4) Liver, renal, heart or lung transplanted patients.
5) Systemic diseases such as Crohn disease or chronic rheumatic diseases receiving immunotherapy, particularly monoclonal antibodies against TNF (195,196).

6) The chemotherapy regimen : regimens including corticosteroids, anthracyclines (191,194), and more recently regimens containing Rituximab (anti-CD20) plus steroïds (192).

Management of HBV reactivation

Lamivudine at the dose of 100 mg daily is currently the only recommended treatment (190,191).

The efficacy of the antiviral therapy administered after symptoms have developed, has yet to be demonstrated (191). In hepatitis following HBV reactivation, mortality has been reported in up to 40% despite treatment with lamivudine (190). It has been postulated that this is due to a delay in antiviral administration at a time when severe hepatic impairment has already occurred (190, 191).

A large body of clinical trials strongly supports the efficacy of pre-emptive therapy in patients undergoing chemo- or immunotherapy (197,198). The review of 12 prospective trials of pre-emptive therapy with lamivudine including 173 treated patients and 116 controls has shown that the rate of hepatitis ranged from 0-20% (mean 9,2%) in lamivudine-treated patients versus 33-67% (mean 54%) in controls. Similarly, the rate of HBV reactivation ranged from 0-24% (mean 8,7%) in lamivudine treated patients to 29-56% (mean 37%) in controls (197).

When starting the preventive treatment ? It is generally recommended to start pre-emptive antiviral therapy one week before the initiation of chemotherapy (190,191). However, if the viral load is higher than 2000 IU/mL, it could be wise to start chemotherapy only when the viral load is under 2000 IU/mL (193).

When stopping pre-emptive antiviral treatment ? There is a real risk of HBV reactivation after withdrawal of pre-emptive lamivudine and the optimal duration for prophylaxis remains unclear (197). In a series of 46 patients with haematological malignancies, 11 (24%) developed HBV reactivation after pre-emptive lamivudine was withdrawn 3 months after discontinuation of chemotherapy, and 3 developed fulminant hepatitis (193). The main predictive factor was pre-chemotherapy HBV DNA $\geq$ 2000 IU/ml (RR 17.82, 95% CI : O.56-37.78). So, it could be advisable to stop pre-emptive antiviral treatment 3 months after cessation of chemotherapy in patients with pre-chemotherapy viral load $\leq$ 2000 IU/mL and to continue pre-emptive treatment at least 1 year in patients with a viral load > 2000 IU/mL.

Recommendations to prevent HBV reactivation in immunosuppressed patients

- *All patients who are candidates for chemo- or immunosuppressive therapy should be screened for HBV markers and all naïve patients should be immunized against HBV as soon as possible (evidence level II-3).*
- *Patients who are HBsAg positive should be offered pre-emptive treatment with lamivudine at 100 mg/day regardless of their HBeAg or HBV DNA status. In these patients, lamivudine should be started at least 1 week before initiation of chemotherapy and should be continued for at least 3 months after discontinuation of chemotherapy. In case of pre-chemotherapy HBV DNA > 2000 IU/mL, immunosuppressive chemotherapy should preferably be started when viral load has declined under 2000 IU/mL and antiviral treatment should be continued for at least 1 year (evidence level III).*
- *Patients who are HBsAg negative but positive for anti-HBc alone with HBV DNA detectable (occult HBV infection) should be treated similarly to patients positive for HBs Ag (evidence level III).*
- *Patients who are HBsAg negative, anti-HBc positive, anti-HBs positive (resolved infection) should not be treated prophylactically but should be monitored for HBsAb, ALT elevation and HBV DNA rise (every 2 or 4 weeks) during chemotherapy and 3 to 6 months after stopping immunosuppressive treatment (evidence level III).*

X.4. *HBV and liver transplantation*

The risk of HBV reinfection after orthotopic liver transplantation (OLT) is approximately 80% and is related to the level of HBV replication at time of transplantation (199). In the early 1990s, reinfection by HBV was associated with 2-year graft and patient survival of 50%, compared to 80% in those patients transplanted for other types of liver disease (199,200). The risk is particularly high when HBV DNA is > 20,000 IU/mL at transplantation. Liver transplantation for HBV-related liver disease has changed from a contraindication to outcomes comparable with non-HBV-related liver transplantations. This dramatic policy change results from the successful implementation of hepatitis B immunoglobulins (HBIg) and antiviral agents aiming at inhibition of HBV-replication before, during and after OLT. Despite the fact that the risk of reinfection has been reduced to 10% or less with current strategies (201), the treatment of a patient with HBV infection and who is a potential liver transplant candidate (HCC, decompensated liver disease), remains a challenge that should be discussed with a liver transplant center.

X.4.1. Control of HBV infection before OLT

When a patient is on the waiting list for OLT – irrespective of the indication – HBV DNA should be checked and if detectable, viral eradication should be pursued. The antiviral strategy used to control the viral load is a matter of debate, which is influenced by previous antiviral therapies, the existence of mutant strains and the waiting-time foreseen. The importance of choosing antiviral therapy with the goal of maximizing viral suppression and minimizing the risk of drug resistance is especially critical in this patient population. It should be aimed at reducing HBV DNA to < 10^{2-3} copies/mL $\approx$ 20-

200 IU/mL) (201). In case of an anticipated short waiting time (< 6 months) lamivudine therapy is acceptable, as there is a limited risk for YMDD-mutations. It should be stressed that for patients with advanced liver failure, it is important not to wait for the clinical benefit of an antiviral effect before listing a patient for OLT. From 6 months on, the addition of adefovir is pivotal to minimize the occurrence of mutant strains. For patients with lamivudine-resistant HBV and elevated HBV DNA levels, addition of adefovir is recommended to inhibit viral replication. In patients with high MELD-scores, mainly because of renal insufficiency, the dose of antiviral agents should be carefully adapted to kidney function (201).

Patients who are HBsAg-positive before liver transplantation, but who are HBV DNA negative, should not be treated with antiviral drugs before OLT.

X.4.2. Prevention of HBV reinfection during and after OLT

Patients who are on an antiviral agent at the time of liver transplantation, should continue this therapy indefinitely.

In addition, all patients who are HBsAg-positive at the time of liver transplantation – irrespective of their replicative status – should receive immunoprophylaxis with HBIg intravenously (IV). However, the dose of HBIg and the target titer of hepatitis B surface antibody remains partially unclear. It is common practice to give HBIg at doses necessary to maintain anti-HBs titers > 500 IU/L in the first week after OLT, which consists most often of 10,000 IU of HBIG given during the anhepatic phase and then daily for the first 8 postoperative days. For patients with active replication at the time of transplantation (> 20,000 IU/mL), anti-HBs levels > 500 IU/L are recommended for the first month, > 250 IU/L to third month and possibly longer, and > 100-150 IU/L thereafter (201). For patients without active replication at the time of OLT target levels of anti-HBs of 100-150 IU/L are probably sufficient, following the highly dosed first week schedule IV.

All patients should regularly (every 3 months) be monitored for breakthrough HBV infections (either reinfection or emergence of viral resistance).

X.4.3. Treatment of HBV infection after OLT

The transplant population with active HBV replication is a heterogeneous group of patients including (a) recurrent HBV infection due to absence (rare) or failure of prophylaxis with HBIg and/or antiviral agents, and (b) *de novo* HBV infection.

Frequently there is a high level of HBV-replication and a rapid evolution of histological disease in the setting of ongoing immunosuppressive treatment. Selection of the optimal therapy for these patients depends on the treatments previously received and the presence or absence of drug-resistant HBV species.

Lamivudine is effective in the absence of YMDD-mutants : several studies (> 200 patients) showed suppression of HBV-DNA to undetectable levels in 68% to 100% of patients treated for periods of 12 to 36 months. As expected, prolonged therapy (> 6 months) was associated with the emergence of YMDD-mutants in 14-62% of patients (201,202). In case of resistance, adefovir should be added. Adefovir has recently been found safe and effective in more than 75% of wait-listed or post-transplantation patients with lamivudine-resistant HBV and prevents graft reinfection with or without HBIg (118). Entecavir is not an ideal rescue therapy for patients with lamivudine-resistant HBV (7,203,204). Interferon is not very efficient in this setting and there is a potential risk of precipitating rejection of the graft (205).

Combination therapy

Although both HBIg and lamivudine are efficacious in preventing HBV recurrence after liver transplantation, recurrence rates in low risk patients at a long term may be as high as 30% and 50 % in patients receiving HBIg or lamivudine as monotherapy, respectively. Therefore, most of the centres prefer a combination of both drugs (206). With this combined approach, HBV recurrence rates have been reduced below 10% (201). In principle, prolonged (life-long ?) prophylactic treatment should be maintained in immunosuppressed patients because HBV DNA has been detected in the serum, peripheral blood cells or the liver even in HBsAg-negative patients 10 years after liver transplantation (207). Given the high cost of the combination approach, current research is devoted to determining whether both molecules need to be given on a life-long basis. Furthermore, efforts are being made to examine whether and when after liver transplantation patients with a lower risk of reinfection such as patients superinfected with the hepatitis D virus, transplanted for acute liver failure or HBV DNA negative at the time of transplantation (207) could benefit from alternative strategies (e.g. vaccination) (201). Nevertheless, one has to keep in mind that more than 50 % of patients with HBV DNA levels greater than 100,000 copies/mL (≈ 20,000 IU/mL) at the time of transplantation develop graft reinfection thereafter (208).

Recommendations for the pre-, per- and posttransplantation period

- *<u>Pre-transplant HBV management</u> (evidence level II-1, III)*
  - *At the time of OLT, the HBV DNA should be as low as possible*
  - *If the waiting time is < 6 months, lamivudine therapy is acceptable*
  - *If the waiting time is > 6 months, the most potent available compound for suppressing HBV DNA associated with the lowest resistance rate is pivotal in order to minimize the occurrence of mutant strains.*

– *In the case of lamivudine-resistant HBV and elevated HBV DNA levels, addition of adefovir is recommended.*
– *If HBsAg-positive before liver transplantation but with negative HBV DNA, no treatment is needed pre-OLT.*

Per- and early posttransplant HBV management : preventing reinfection
– *All patients HBsAg-positive at the time of OLT (irrespective of their replicative status) should receive immunoprophylaxis with HBIg (evidence level II-1).*
– *10,000 IU of HBIg is generally given during the anhepatic phase and then daily for the first 8 post-operative days, further doses of HBIg depend on the anti-HBs titer that is aimed at.*
– *Recommended anti-HBs titers (evidence level III) :*
  • *anti-HBs titers > 500 IU/L in the first week after OLT*
  • *when active replication at the time of OLT, anti-HBs levels > 500 IU/L are recommended for the first month, > 250 IU/L to third month and possibly longer, and > 100-150 IU/L thereafter*
  • *When no active replication at the time of OLT target levels of anti-HBs of 100-150 IU/L are probably sufficient, following the highly dosed first week schedule.*
– *All patients should regularly (every 3 months) be monitored for breakthrough HBV infection .*

• *<u>Post-transplant HBV management</u> (evidence level II-1, III) :*
– *Patients on antiviral agents at the time of OLT, should continue this therapy indefinitely together with HBIg.*
– *Combination therapy of HBIg + antiviral agent should, currently, be continued lifelong.*

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