Antiviral treatment for chronic hepatitis B

CL Lai, PC Wu

An updated review of the antiviral agents currently available or under trial for the treatment of chronic hepatitis B is presented. There are two broad groups: (1) immunomodulators including interferon α (which also has a direct antiviral effect), thymosin α_1 and Theradigm-HBV and (2) viral suppressors such as famciclovir and lamivudine. These agents are still in clinical trial worldwide, singly or in combination. Their long term efficacy in the treatment of hepatitis B remains to be evaluated.

Introduction

It is estimated that as many as 25% to 40% of the 300 to 350 million hepatitis B surface antigen (HBsAg) carriers in the world will eventually die from cirrhosis of the liver and/or hepatocellular carcinoma (HCC).[1] Since there is a progressive increase in the incidence of HCC with age, it has been suggested that all HBsAg carriers will eventually die from HCC and/or cirrhosis if they live long enough.[2]

The ultimate aim in the treatment of HBsAg carriers is therefore to decrease, or prevent altogether, the development of cirrhosis and HCC. This requires decades of follow up of treated patients. More realistic short term objectives (usually used in clinical trials) include the following: (1) viral suppression, which is established by the disappearance of serum hepatitis B virus (HBV) DNA (using hybridization assay) and hepatitis Be antigen (HBeAg) with or without antibody against HBeAg (anti-HBe); (2) decreased liver damage, established by the normalisation of serum transaminase levels (if these were previously elevated) and improved liver histology; and (3) complete eradication of HBV, this is indicated by the loss of HBsAg and detectable HBV DNA in the serum and liver, even by polymerase chain reaction (PCR) assay or branch DNA chemiluminescent amplification technique. This last objective is seldom achieved with the current treatment regimens.

Department of Medicine, Queen Mary Hospital, Pokfulam, Hong Kong
CL Lai, MD, FRCP
Department of Pathology, The University of Hong Kong, Pokfulam, Hong Kong
PC Wu, MD, FRCPath

Correspondence to: Prof CL Lai

Table 1. Agents currently used or under trial for treating chronic hepatitis B

Immunomodulators
Interferon α
Thymosin α_1
Therapeutic vaccines, eg, Theradigm-HBV

Viral suppressors
Famciclovir
Lamivudine
Ganciclovir

The agents currently used or under trial for the treatment of chronic hepatitis B can be broadly separated into two groups (Table 1). The first group are the immunomodulators, which act by modulating the immune response of the host to the HBV antigens that are expressed on the surface of the hepatocytes.[3] The main mode of action of interferon alpha (IFNα) in HBsAg carriers is immunomodulation, but it also has a direct antiviral effect. Other newer immunomodulators include thymosin α_1 (Tα_1) and therapeutic vaccines, such as Theradigm-HBV (Cytel, San Diego, Ca, US). The second group of agents comprises viral suppressors. The most promising agents in this group are famciclovir and lamivudine (Glaxo Wellcome, London, UK).

Interferon alpha

This is currently the only agent approved for use in the treatment of HBsAg carriers. It is given subcutaneously at a dose of 5 mU daily or 10 mU thrice weekly for 16 weeks. A meta-analysis of 15 randomised placebo-controlled studies showed that IFNα

Table 2. Summary of the results of a meta-analysis of 15 randomised placebo-controlled trials of interferon alpha in chronic hepatitis B[4]

	IFNα[*] treated group (%)	Control group (%)	P value
Loss of HBV DNA	37	17	P<0.0001
Loss of HBeAg	33	12	P<0.0001
Loss of HBsAg	7.8	1.8	P<0.001

[*]IFNα interferon alpha

was beneficial (Table 2).[4] Its usefulness, however, is limited. Loss of HBeAg and HBV DNA occurred only 20% more frequently in treated patients than in control patients and loss of HBsAg occurred in only 6% more patients. Interferon alpha had a significant effect on the normalisation of alanine aminotransferase (ALT) levels. Most studies have demonstrated that successfully treated patients have liver biopsies that histologically, show less evidence of damage.

When the cost-effectiveness of IFNα treatment was calculated based on the meta-analysis of nine randomised trials,[5] it was concluded that for a 35-year-old individual with HBeAg-positive chronic hepatitis B, IFNα increases life expectancy by only 3.1 years or 3.4 quality-adjusted life-years. This modest increase in life expectancy was, however, based on certain questionable assumptions. The authors calculated that patients who are HBeAg-positive and HBsAg-positive have a 12.1% probability of developing compensated cirrhosis annually; whereas patients who are HBeAg-negative and HBsAg-positive have only a 1.0% probability of developing the condition. It is now generally accepted that cirrhosis develops as a result of the immunological injury to the liver that occurs during the viral clearance phase when a patient is seroconverting from HBeAg positivity to anti-HBe positivity.[6-8] The assumption therefore that HBeAg-negative patients are much less likely to develop cirrhosis compared to HBeAg-positive patients seems questionable.

Interferon alpha treatment combined with steroid priming

Steroid therapy in a chronic hepatitis B patient causes enhanced viral replication,[9] possibly acting through the glucocorticoid-responsive element in the HBV genome.[10] Following withdrawal of a short course of steroid, there is a decline in HBV DNA and HBV DNA polymerase levels that coincides with an immunological rebound with enhanced T lymphocyte function.[11] This improved T lymphocyte function may potentiate the effect of interferon. A large multicentre trial, however, has shown an almost identical rate of HBeAg seroconversion in patients treated with IFNα alone and in patients pre-treated with steroid followed by IFNα.[12] Moreover, steroid withdrawal is documented to precipitate hepatic decompensation (as a result of hepatocyte necrosis) in patients with marginal liver function, which can be fatal in some cases.[13-15] Such fatal reactivation on withdrawal of steroid can sometimes occur in completely asymptomatic HBsAg carriers[16] and steroid should therefore be used with caution in HBsAg carriers. Withdrawal of steroid is not recommended as a routine procedure prior to a course of IFNα.

Response of Oriental hepatitis B carriers to interferon alpha

At least 50% of Oriental HBsAg carriers acquire the infection perinatally,[17] and a large proportion of the remainder acquire the infection within the first few years of life.[18] Such early infection is believed to induce immune tolerance to HBV, possibly through deletion of T cells that can recognise HBeAg and the hepatitis B core antigen (HBcAg).

Two large scale trials have been carried out in adult Chinese HBsAg carriers in Hong Kong.[19,20] The results of the second trial are summarised in Table 3. In this study, the carriers were randomised into those who had normal pre-treatment ALT levels and those with persistently elevated pre-treatment ALT levels (raised for more than three consecutive months). Interferon alpha had little effect in patients with normal ALT levels, while the response to IFNα, with or without prednisone priming, was slightly better in patients with elevated ALT levels. The elevated ALT in these patients, however, also suggests that they were in the viral clearance phase. The spontaneous seroconversion rate to HBeAg positivity in the control group was as high as 19% within one year. Hence, the authors concluded that even in patients with elevated ALT levels, "it is not clear whether interferon merely

Table 3. Summary of the response to interferon alpha treatment in Chinese adult HBsAg carriers[20]

	Prednisone + IFNα[*] (%)	IFNα alone (%)	Control group (%)	P value
Patients with elevated ALT				
loss of HBeAg	43	33	19	ns
loss of HBsAg	0	6	0	ns
Patients with normal ALT				
loss of HBeAg	0	10	0	ns
loss of HBsAg	0	5	0	ns

ns   not significant

[*]IFNα   interferon alpha

hastened the process of endogenous inhibition of HBV replication or provided actual benefit to some patients."[20]

Two randomised trials have also been carried out in Chinese children who are hepatitis B carriers.[21,22] However, the results have been equally disappointing. Hence, the poor response seen in adult carriers does not appear to be related to delayed treatment of the disease.

Effect of interferon alpha in producing viral eradication

A meta-analysis of 15 randomised trials has shown that loss of HBsAg positivity occurred 6% more often in IFNα-treated patients than it did in control patients, six months after treatment ceased.[4] On long term follow up, 65% of IFNα-treated Caucasian patients who have had a sustained loss of HBeAg also lose HBsAg.[23] However, only 24% of Spanish responders and none of the Chinese responders lost HBsAg on long term follow up.[24,25] Complete disappearance of HBV DNA as tested by PCR assay, occurs only rarely in IFNα-treated patients who clear HBeAg but remain positive for HBsAg. Of the 272 Chinese adult and child HBsAg carriers who participated in three randomised placebo-controlled trials, 5.9% of the treated patients and 1.2% of the control patients became HBV DNA negative on long term follow up, a statistically insignificant result.[26]

Adverse effects of interferon alpha

The adverse effects of IFNα are dose-related and can be severe (Table 4). Although most adverse effects can be decreased by reducing the dosage of IFNα, some can lead to the premature ending of therapy. The initial influenza-like syndrome can be minimised by

Table 4. Adverse effects of interferon alpha

Influenza-like syndrome (rapid tachyphylaxis)
Myalgia
Marrow suppression (usually moderate)
Alopecia
Depression (occasionally severe)
Autoantibody induction
Induction of interferon-neutralising antibodies
In patients with advanced hepatitis/cirrhosis
  - susceptibility to bacterial infections
  - liver function deterioration

gradually increasing the dose. The induction of auto-antibodies, although usually clinically silent, can lead to overt hypo- and hyper-thyroidism and idiopathic thrombocytopenic purpura.[27] Interferon therapy may also induce autoimmune hepatitis in individuals who are genetically predisposed to developing auto-immune liver disease.[28,29] The presence of interferon neutralising antibodies is often of no clinical importance, but when present in large amounts, they can counteract the effects of IFNα. In patients with advanced hepatitis, IFNα may worsen liver function. It also increases susceptibility to bacterial infection, and should therefore be used only under close monitoring in patients with mild or moderate hepatic decompensation.

Thymosin alpha 1

This is an immunomodulator that has several modes of action; it primarily increases the efficiency of T cell maturation.[30] It also acts on T cells after maturation, increasing the production of cytokines such as interferon γ and interleukin 2,[31] and it upregulates the expression of cytokine receptors.[32] A study of

woodchuck hepatitis virus-infected woodchucks treated with Tα_1 seems promising.[33]

In a phase II trial involving patients with chronic hepatitis B, seven patients received Tα_1 while five patients received subcutaneous placebo injections twice weekly for six months.[34] More of the thymosin-treated patients cleared HBV DNA from their sera, and had an absence of replicative forms of HBV DNA in their liver biopsies, compared with patients in the control group (P<0.04). A response to thymosin therapy was associated with improved peripheral lymphocyte, CD3, and CD4 counts, and in interferon γ production. The serological improvements were sustained during follow up.

In an American Phase II, multicentre, randomised, controlled trial involving 99 patients, the preliminary results suggest a trend in favour of Tα_1 compared with placebo, but the results are far less promising than those of the phase II trial.[35] Other phase III trials are being performed in Taiwan and Singapore. Any definitive conclusions concerning the usefulness of Tα_1 must await the final outcome of these phase III trials.

The effect of combining Tα_1 with lymphoblastoid IFN has been tested in 15 patients. Preliminary data show a loss of HBV DNA in 60% of patients and a loss of HBsAg in 40%.[36] Again, firm conclusions have to await the results of further trials. As to the safety of Tα_1, no adverse events have been reported to date in the more than 1000 subjects who have been treated with the drug.

Therapeutic vaccines

Patients who clear HBV develop a strong HLA class I-restricted response; this response is weak in HBsAg carriers. Both withdrawal of steroid and IFNα probably act by inducing cytotoxic T lymphocytes (CTL).

A therapeutic vaccine, Theradigm-HBV, has been designed to induce a CTL response to HBV.[37] Theradigm-HBV has three components: the HBcAg peptide 18-27, and two additional components for enhancing immunogenicity, i.e. tetanus toxoid peptide 830-843. The HBcAg peptide 18-27 was selected because it has been shown to be recognised by CTL from patients with acute HBV infection.[38,39]

A dose escalation trial in 26 normal subjects showed that Theradigm-HBV is safe to use. It induces a primary HBV-specific CTL response with the first dose and a booster effect with the second dose, given 28 to 35 days later. A phase II study is underway to determine the efficacy of vaccinating HBsAg carriers with two doses of Theradigm-HBV.

Famciclovir

Famciclovir is the oral derivative of penciclovir. It is converted to penciclovir in the intestinal wall and in the liver after absorption.[40] Penciclovir is a guanine analogue that inhibits viral DNA chain elongation by competing for viral DNA polymerase in its active intracellular triphosphate form.[41] It has a potent effect on herpes simplex and zoster viruses.[42]

Famciclovir suppresses the replication of the duck hepatitis B virus in the Pekin duck and human HBV replication in human hepatoma cells transfected with the HBV genome.[43] In a double-blind placebo-controlled pilot study, 17 patients were randomised to receive famciclovir 250 mg three times daily, or 500 mg three times daily, or placebo.[44] Of the 11 evaluable patients who received famciclovir, six showed a greater than 90% reduction in their HBV DNA level compared with no reduction in the patients who received placebo. In four of the responders, the fall in HBV DNA was sustained throughout the 10-day treatment, and in two of these, the fall persisted for a further two weeks. More prolonged courses of famciclovir are now being assessed for its effect on HBV replication.

Famciclovir has been found to be remarkably safe in patients with herpes zoster and genital herpes.[42] The most common adverse effects of headache, nausea, and diarrhoea occur with equal frequency in patients receiving famciclovir and those receiving placebo.

Lamivudine

Lamivudine [the (-) enantiomer of 2´ deoxy-3´-thiacytidine] is a 2´3´ dideoxynucleoside similar in structure to 3´-deoxy-3´-azidothymidine (AZT), 2´3-dideoxycytidine (ddC), and 2´3-dideoxyinosine (ddI), drugs used for the treatment of patients with human immunodeficiency virus (HIV) infection. These agents inhibit DNA synthesis by causing chain termination of the nascent proviral DNA in addition to interfering with the reverse transcriptase activity of HIV and HBV. Lamivudine was approved for use in HIV patients in late 1995 because of its synergistic effect with AZT.[45] Lamivudine inhibits the replication of HBV in human HBV-transfected cell lines[46] but replicative HBV DNA reappears when it is withdrawn.[47] It is also very effective in vivo when given to infected ducks and chimpanzees.

Multiple phase II dose-ranging trials with or without placebo have been performed in both Caucasian and Oriental HBsAg carriers.[48-50] Oral daily doses of 5 mg to 600 mg were given for 4 to 12 weeks. The results of these trials are essentially identical; HBV DNA dropped to very low levels within one week of therapy. The suppression of HBV DNA was suboptimal with daily 25 mg or smaller doses, but 100% of patients receiving 100 mg or greater doses daily had almost complete suppression of HBV DNA (95% to 100% of pre-treatment levels). There were decreases in HBeAg and HBsAg titres (Glaxo, unpublished data) and normalisation of ALT levels.[49] These occurred irrespective of the pre-treatment HBV DNA and ALT levels in both Caucasians and Orientals.[48,49] In the majority of patients, HBV DNA, HBeAg, and HBsAg, returned to pre-treatment levels 4 to 8 weeks after treatment. In 19% of the patients who were given lamivudine for 12 weeks, however, HBV DNA suppression was sustained.[49] The HBeAg disappeared in 12% of patients.

Several large scale multicentre controlled trials are now being performed in Southeast Asia, Europe, and the United States, comparing lamivudine with or without IFN, and with placebo. Since the phase II trials show that the majority of patients have a rebound of the HBV DNA on cessation of therapy, lamivudine probably has to be given on a long term basis. Suppression of viral replication per se may not be sufficient; there must be a corresponding decline in the number of productively infected hepatocytes.[50] Reduction in the number of infected hepatocytes depends on the spontaneous death of infected hepatocytes and probably also on the inefficient passage of covalently-closed circular DNA (ccc DNA) of HBV from infected hepatocytes that proliferate to replace dying cells.[51] Viral suppressors may therefore have to be administered over many months, if not years. Patient compliance with oral lamivudine taken once daily should be good. One conceivable endpoint for the cessation of therapy would be the complete eradication of HBV, i.e. the disappearance of HBsAg and HBV DNA from the serum and liver, as tested by PCR assay.

It is potentially dangerous to stop lamivudine prematurely. One report describes a rise in ALT that occurred in a patient four weeks after discontinuing lamivudine.[52] This rose to 100 times the upper limit of normal four months later, with the appearance of jaundice; a liver biopsy showed necrosis of hepatocytes. The authors postulate that a reinfection of hepatocytes occurred after the lamivudine was with-drawn, followed by a severe immune reaction, as if the patient were experiencing an acute hepatitis B episode. The patient survived, with clearance of HBV DNA and HBeAg after a course of prednisone. The authors documented that such a rise of ALT occurs in 16% of patients within 8 to 24 weeks of the stopping of lamivudine treatment. This report supports that lamivudine may have to be maintained on a long term basis until there is evidence of complete viral eradication.

Lamivudine is otherwise a remarkably safe drug, unlike fialuridine, an investigational drug that caused severe hepatic failure and lactic acidosis in seven of 15 patients, five of whom died.[53] The toxicity of fialuridine is probably related to its free 3´-hydroxyl group that allows its incorporation into DNA, including mitochondrial DNA, at internucleotide linkages.[54] Lamivudine lacks this free 3´-hydroxyl group.

Compared with other non-fialuridine nucleoside analogues, lamivudine also has fewer potential side effects. Drugs like AZT can cause myopathy and marrow hypoplasia.[55,56] It has been shown in rats that AZT (which is not incorporated into mitochondrial DNA) inhibits mitochondrial DNA, RNA, and polypeptide synthesis.[55] In contrast, lamivudine, when tested in cell cultures, has much less effect on mitochondrial DNA content and on the activity of DNA polymerase γ, the enzyme responsible for the replication of mitochondrial DNA.[43]

The adverse events that have been encountered with lamivudine in clinical settings are summarised in Table 5. These events were reported for a total of 19 520 patients, of whom 631 had HBV infection and 18 889 had HIV infection. A large proportion of the latter patients had been receiving lamivudine for more than two years.

The haematological abnormalities include mild anaemia, neutropenia, and thrombocytopenia. A haematogical abnormality was reported in only one patient with HBV infection, and that patient had a liver transplant. Neurological events were mostly in the form of muscle weakness. Some patients developed elevations of creatine kinase levels but most of these patients were symptom-free.

Pancreatitis has so far been reported only in patients with HIV infection. Approximately one third of the patients who developed pancreatitis had had pancreatitis prior to lamivudine treatment. It was often difficult to decide whether the pancreatitis was causally

Table 5. Adverse events related to lamivudine consumption observed in 19 520 patients[*]

	No. of patients with HIV infection	No. of patients with HBV infection	% of total patient population
No. of patients	18 889	631	
Nature of adverse events encountered			
Haematological	103	1	0.53
Neurological	54	0	0.28
Pancreatic	52	0	0.27
Other hepatobiliary complications	23	5	0.14
Gastrointestinal	39	0	0.20
Endocrineal	26	2	0.14
Musculoskeletal	8	1	0.05
Psychiatric	3	-	0.02

[*]Data kindly supplied by Glaxo Research and Development Ltd.

related to lamivudine, a complication of HIV infection, or whether it was due to concomitant drug treatment (e.g. steroids and AZT).

Other hepatobiliary complications included elevated transaminase (not related to withdrawal of lamivudine), bilirubin, lipase, and amylase levels. These patients were mostly symptom-free. A small proportion of patients had diarrhoea, nausea, hypoglycaemia, and myalgia. In all, only 1.6% of patients were reported as having complications, most of which were mild. Lamivudine is a safe drug to use in patients with chronic hepatitis B.

Viral suppressors in liver transplantation

Hepatitis B immune globulin (HBIG) is the only agent proven to be of use for the prevention of HBV recurrence after liver transplantation for end-stage hepatitis B cirrhosis. In a study of 372 HBsAg-positive patients by Samuel and colleagues,[57] the three-year actuarial risk of recurrence of HBV infection was 83% for HBV DNA-positive patients, 66% for HBV DNA-negative, HBeAg-positive patients, and 58% for patients negative for both HBV DNA and HBeAg. With long term administration of HBIG, the three-year risk of recurrence was significantly reduced (P<0.001) but was still 36%. The dosing of HBIG is difficult, as it requires constant monitoring of anti-HBs titres. Its cost is also considerable and is estimated to be US$30 000 per year per patient.

Recently, more encouraging results have been achieved with the three viral suppressors, ganciclovir, famciclovir, and lamivudine. The first two agents have been given to patients who had recurrent HBV infection despite HBIG treatment. In one series, ganciclovir was given intravenously to eight of these patients (and to one patient with de novo HBV infection) for three to 10 months.[58] In all patients, serum HBV DNA levels fell (mean decrease = 90%). The ALT levels also decreased. Hepatic expression of HBV antigens and HBV DNA was reduced in three of six patients assessed. After discontinuation of ganciclovir, HBV DNA rebounded in all patients with the exception of the patient with de novo infection. Four patients underwent re-treatment with success.

Kruger et al used famciclovir (given orally) in 38 patients with HBV re-infection following liver transplantation.[59] There was a reduction in serum HBV DNA (median 91% decrease) in 83% of evaluable patients. Eight patients were given famciclovir for more than one year without significant side effects.

Lamivudine has been used as a prophylactic agent to prevent hepatitis B re-infection following liver transplantation. It is given orally as a single agent, without HBIG, for at least four weeks prior to transplantation. In one of the first published abstracts, 12 patients were transplanted, with one early post-operative death.[60] Before transplantation, all patients were found positive for HBV DNA by PCR assay. After transplantation, only one patient had recurrent HBV infection. The 10 surviving patients all became negative for serum HBV DNA and for HBsAg and HBcAg in the liver biopsies. Six of these patients also lost HBsAg. The drug was well tolerated except in one patient who developed sensorimotor neuropathy and myopathy.

Viral suppressors thus appear to provide an effective alternative to HBIG in the prevention and treatment of HBV re-infection after liver transplantation in HBsAg-positive subjects.

Conclusion

For the treatment of chronic hepatitis B, IFNα is only of limited use, especially in Oriental populations. Newer immunomodulators such as Tα_1 and therapeutic vaccines are being assessed clinically. So far, Tα_1 appears to generate only a modest effect.

Several viral suppressors of HBV replication have been assessed. Of these, lamivudine has proved to be the most potent. However, viral suppressors probably need to be taken on a long term basis to achieve an effective decline in the number of productively infected hepatocytes.

Viral suppressors have also been found to be useful in the treatment and prevention of recurrence of HBV after liver transplantation for end-stage hepatitis B liver disease. When given before liver transplantation, a significant proportion of patients become HBsAg-negative after the transplantation.

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