# Long Term Low Dose Interferon Alpha-2b in the Treatment of Chronic Hepatitis B in Multi-Ethnic Patients in Hawaii

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*The antiviral and immunomodulatory effects of interferon were assessed in the treatment of chronic hepatitis B in multi-ethnic patients to prevent viral replication and chronic liver damage. Five million units of recombinant interferon alpha-2b were administered three times a week for 48 weeks to a group of 18 chronic active hepatitis B patients. A complete response was defined as seroconversion to anti-HBe and/or loss of HBe antigen. Seroconversion to anti-HBe in 5 of 12 (42%) chronic active hepatitis B patients occurred after 48 weeks of therapy. HBV-DNA decreased to undetectable levels in 8 of 12 (67%) patients. This chronic low-dose interferon administration regimen demonstrated responses comparable to other studies.*

## Introduction

Chronic hepatitis B is a major public health problem in Hawaii because of the large immigrant population from Asia and the Pacific Basin. Exposure to hepatitis B virus (HBV) often results in chronic hepatitis that can significantly increase the risk of developing cirrhosis and hepatocellular carcinoma.[1] Approximately 30,000 to 40,000 in Hawaii have been infected with hepatitis B virus (HBV). The estimated carrier rate in Hawaii is 2-3%, higher than the 0.5% of the mainland United States.[2] In acute HBV infection in adults, 90% of those infected will develop a primary antibody response; 5 to 10% of patients will develop chronic inflammation.[3] More than 90% of neonates born to HBeAg-positive mothers have been reported to develop chronic HBV infection.[4,5] Interferon has been shown currently to be the most effective therapy in approximately 30 to 40% of chronic hepatitis B patients[6,7,8] because of its antiviral and immunomodulatory effects. Perillo et al[8] have reported hepatitis Be antibody (anti-HBe) seroconversion and loss of HBV DNA in patients treated with interferon with (36%) and without (37%) prednisone. Long term remission with formation of anti-hepatitis Bs antigen (anti-HBs) was observed by Korenman et al[9] in patients followed for 3 to 7 years after interferon alpha-2a therapy. This represents a hopeful therapeutic option for patients who are not able to spontaneously suppress the HBV infection immunologically.

Based on our previous experience with interferon protocols in selected cancer patients,[10] the daily administration of 5 million units interferon daily over 16 weeks as utilized by Perillo et al[8] would not be a tolerable dose for our population in Hawaii because of the constitutional symptoms of interferon. In our cancer patients, we first used a lower interferon dose, 1 million units, administered three times a week because the rest periods made this therapy more tolerable and natural killer cell function was better maintained.[10,11] The majority of our chronic hepatitis B patients were fairly asymptomatic before therapy and gainfully employed. Fatigue and weakness resulting from the medication would preclude many from continuing their occupations.

A Phase II protocol was initiated to treat chronic hepatitis B patients to determine the effect of 5 million units of recombinant interferon alpha-2b (Schering-Plough Corporation, Kenilworth, NJ) administered subcutaneously three times a week for a period of 48 weeks. The objective was to utilize the antiviral and immunomodulatory effects of interferon to prevent viral replication and the chronic liver damage with the increased risk of developing hepatocellular carcinoma. These studies indicated that this interferon dose and schedule could suppress HBV replication and result in HBeAg seroconversion and normalization of biochemical liver function activity.

## Materials and Methods

### Patient Population

Adult volunteers were recruited from the database of hepatitis B carriers maintained by the Hawaii Department of Health.[12] These patients were re-staged for HBV serological markers: hepatitis B e antigen (HBeAg), antibody to HBeAg (anti-HBe), hepatitis B surface antigen (HBsAg), antibody to HBsAg (anti-HBs) and serum alanine aminotransferase activity (ALT), to determine the stage of their hepatitis B infection. From this database, 17 of 95 (18%)

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patients screened had chronic active hepatitis B: HBsAg and HBeAg positive; ALT levels were 100 IU/L ± 128. Gastroenterologists also referred patients for evaluation and therapy.

Chronic active hepatitis patients, HBeAg and HBsAg positive for greater than 6 months, anti-HBe and anti-HBs negative, ALT levels >1.3 x high normal (45 IU/L), were selected for therapy.

### Interferon Therapy

Eighteen chronic active hepatitis B patients were treated with recombinant interferon alpha-2b (Intron-A, Schering-Plough Corporation, Kenilworth, NJ). Three million units were adminstered subcutaneously 3 times per week for 2 weeks and, if tolerated, the dose was increased to 5 million units for a total of 48 weeks.

Patients received their injections in the Ambulatory Oncology Clinic or chose to voluntarily self-administer their medication after training by the Oncology Nursing Staff. The dose was reduced to 3 million units when platelets were <100,000 or granulocytes were <1000. All patients signed written informed consent for treatment with this interferon protocol approved by the Institutional Review Board.

### Evaluation During Therapy

Patients were evaluated during therapy for hematological, biochemical and serological profiles. Blood was collected for complete blood and platelet counts; liver function tests including serum alanine and aspartate aminotransferase and gamma glutamyl transpeptidase activities (ALT, AST, GGPT) prior to therapy, after 2 weeks, monthly during therapy and 2-3 months post therapy. The serological markers for HBV, HBeAg/anti-HBe and HBsAg/anti-HBs, were evaluated at baseline, after 6 months, at the completion of therapy, and 2-3 months post therapy by the Enzyme Immunoassay (Abbott Laboratories, Abbott Park, Il). All evaluations were performed by the same Clinical and Reference Laboratories.

A complete response was defined by loss of HBeAg and/or the formation of anti-HBe or anti-HBs; non-responders showed persistence of HBeAg. A clinical response was defined as the normalization of serum ALT levels.

### HBV DNA Assay

A sensitive and quantitative radiological molecular solution hybridization assay kit for HBV-DNA generously supplied by Abbott Laboratories (Abbott Park, IL) was used for evaluating viral replication and monitoring antiviral therapy.[13] Aliquots of serial serum samples from each patient stored at -70C were tested for HBV DNA at baseline, 2-3 months intervals, the end of therapy and 2-3 months following therapy. The assay utilizes a single-stranded I-labeled DNA probe complementary to HBV DNA sequences and with a minimum specific activity of 10 cpm/μg. Sensitivity of the assay was 1.5 pg HBV DNA per ml of serum or approximately 4.5 x 10E5 HBV genomes per ml serum.

### Statistical Analysis

Results are expressed as arithmetic mean ± S.D. except where noted. Data was analyzed with the Sigma Stat program (Jandel Scientific, San Rafael, CA). Continuous variables were analyzed by linear regression and/or the non-parametric Kruskal-Wallis technique. The IBM 55SX and PS80 PC computers were used.

## Results

### Study Population

The average age of the chronic active patients was 33 ± 10 yrs; 10 of the 18 (56%) were males. Baseline liver function tests (ALT, AST and GGPT) were increased in the chronic active patients: ALT = 96 ± 74 IU/L, AST = 62 ± 39 IU/L, GGPT = 141 ± 304 IU. Immigrants from Vietnam represent the major ethnic group of patients (39%); Filipinos, 22%; Chinese, 17%. All were gainfully employed, housewives or students except for three unemployed medicaid recipients.

### Toxicity

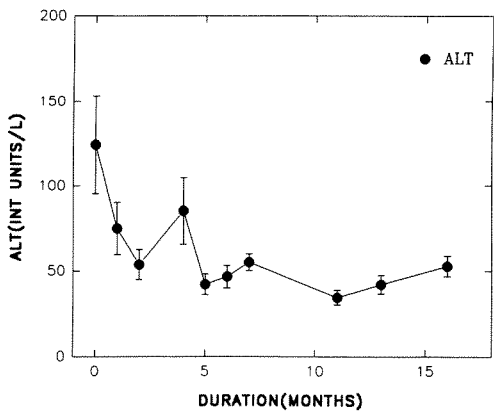
With this interferon protocol at a reduced dose of 5 million units three times a week, 6 of 18 (33%) patients ceased therapy on their own choice. Their reasons included: inability to care for family(2) or to tolerate the side effects.(2) Two gave no reason for terminating treatment.

Although we used a lower dose IFN protocol, 3 of 18 (17%) chronic active patients required dose reduction from 5 to 3 million units because of intolerable constitutional symptoms. In two patients, platelets decreased below 100K. One patient on Dilantin required a dose reduction to 1.5-2 million units because of granulocytopenia.

### Biochemical Liver Function Tests

Assessment of biochemical studies for liver function revealed increased ALT levels in the chronic active hepatitis patients which returned to normal levels in those completing 48 weeks of therapy, $p < .05$ by ANOVA analysis (Figure 1). This occurred whether or not seroconversion occurred. The non-responders had flares of ALT levels after discontinuation of interferon therapy.

Fig 1.—Changes in ALT levels of chronic active hepatitis B patients on interferon therapy, X ± SD. Elevated levels at baseline decreased to normal levels in those patients completing the 48 weeks of therapy (P<.05).




### Serology of HBV Markers

Serological markers for HBsAg/anti-HBs and HBeAg/anti-HBe were assessed to determine the outcomes. Loss of HBeAg and seroconversion to anti-HBe occurred in 5 of 12 (42%) patients after interferon therapy with no relation to the dose received. One patient responded after only six months of therapy; a sixth patient was HBV DNA and HBeAg negative after six months of therapy but did not form anti-HBe. In chronic active patients requiring the full 48 weeks of therapy, antibodies to HBeAg were not detected until 2-3 months post therapy. Seroconversion of HBsAg was not observed. Of the six responders, five were U.S. citizens and there was one female immigrant. Sex was equally distributed.

The remaining six patients were non-responders (HBeAg+, BHeAb-). Follow-up studies will be continued. The ethnicity of the six non-responders of the chronic active hepatitis B patients were Vietnamese (2) and Chinese (2) immigrants, and Filipino (2) born in Hawaii. The six who ceased therapy were not evaluable and were also immigrants, Vietnamese (5) and Chinese (1).

### HBV-DNA

Ten of the twelve patients had elevated levels of HBV-DNA before therapy. All six responders had no detectable HBV-DNA after treatment. Two of the six non-responders did not have measurable levels of HBV-DNA after therapy although no HBV antibodies were formed.

## Discussion

In this study with multi-ethnic patients, we observed the antiviral effect of interferon in 67% chronic active hepatitis B patients who showed progressive decline in HBV DNA during therapy. We achieved a 42% HBeAg seroconversion and HBeAb formation rate in patients who completed the 48-week interferon course. This Phase II protocol of 5 million units interferon alfa-2b administered subcutaneously three times a week for 48 weeks to patients with hepatitis B infection yielded comparable results as reported by Perillo et al[8] using 5 million units daily for 16 weeks and including pretreatment with prednisone. HBeAg seroconversion occurred in 30 to 40% of their patients with loss of HBV DNA in 40 to 50%. Similar rates of seroconversion were reported by Reichen et al[14] with either 1.5 or 5 million units interferon alpha 2b three times a week for 4 months with and without a preliminary course of prednisone therapy. HBsAg seroconversion was not observed in our studies although Korenman et al have reported that anti-HBs may appear at an average of 3 years after treatment.

We have used an interferon dose and schedule of 5 million units three times a week subcutaneously over a longer period of 48 weeks with minimal effect on blood counts. This Phase II protocol would allow greater tolerance of this medication in chronic hepatitis B patients. After approximately 9 months, the cumulative dose of interferon would be equivalent to the daily administration of 5 million units for 16 weeks as used by Perillo et al.[8] Furthermore, patients treated in an outpatient environment would be able to continue their productive work to minimize economic loss.

With this interferon dose, however, six (33%) Asian immigrant patients still discontinued treatment. The constitutional symptoms were most disturbing to these patients who were essentially asymptomatic before therapy. Some were unable to continue working. Our cancer patients have tolerated higher doses but may be more motivated to continue than the chronic hepatitis B patients.

Hawaii has a large percentage of affected Asian-Pacific patients because of the large immigrant population.[2,12] The Asian male has been observed to be the most resistant to treatment.[15] Lok et al[16] reported spontaneous seroconversion in 31 of 142 (22%) HBeAg positive Chinese patients observed up to 60 months. They predicted a 17% one year seroconversion rate. However, none of our Asian non-responders demonstrated seroconversion one year after completion of interferon therapy.

In this study, most of our non-responders were Asian male immigrants; male responders were USA or Hawaii-born. Three female responders, two Hawaii-born and a Filipino immigrant had progressive decline in HBV DNA after interferon therapy. A sustained therapeutic effect in these young females may prevent the vertical transmission of HBV to their newborn and subsequent chronic antigenemia.

We monitored the antiviral effect of interferon by serial quantitation for HBV-DNA using the solution hybridization assay. Eight of twelve (67%) chronic active hepatitis patients had undetectable levels of HBV DNA after the end of therapy. An antiviral effect of interferon is indicated with no seroconversion, as yet, in 2 non-responders. Follow-up studies are planned in these non-responders with no detectable HBV DNA.

Our diverse group of Asian patients represents a major public health problem and challenge for developing effective therapeutic interventions in Hawaii. The possibility exists that these patients may have had prolonged infection since childhood resulting in the transcription of virus to their hepatocytes and may be more resistant to interferon therapy. The elimination of chronic HBV infection is greatest if treated relatively early when viral HBV DNA exists in the non-integrated or episomal state.[17] With the high carrier rate in Hawaii, early detection and treatment as well as vaccination will be necessary for the prevention of the sequelae of chronic HBV infection.

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