Digestive Diseases and Sciences, Vol. 48, No. 1 (January 2003), pp. 140–145 (© 2003)

Intensive Interferon Therapy Does Not Increase Virological Response Rates in African Americans with Chronic Hepatitis C

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To determine if an intensive regimen of daily, high-dose interferon would improve the initial response rates to therapy for hepatitis C genotype 1 among African American and Caucasian patients, we conducted a retrospective analysis of a treatment trial conducted between October 1995 and June 1997. Patients were randomized to 24 weeks of therapy with interferon $-\alpha$-$_{2b}$ at either 5 MU *daily* or 3 MU three times a week. On the standard interferon regimen (3 MU three times a week) African Americans and Caucasians had similar initial response rates. However, unlike Caucasians, African Americans did not have an increased initial virological response when treated with an intensive, daily dose regimen. Levels of HCV RNA decreased more slowly during the first 12 weeks of therapy among African Americans. Nelson-Aalen cumulative hazard estimates for the different race and dose combinations revealed that Caucasians who received daily interferon were most likely to have an initial response (logrank, $P < 0.001$).

KEY WORDS: hepatitis C; interferon; therapy; race; African American.

Major advances have been made over the last decade in the field of antiviral therapy for chronic hepatitis C. Combination therapy with interferon and ribavirin has improved virological sustained response rates to nearly 40% compared to only 10–15% for patients treated with interferon alone. Unfortunately, response to therapy is not uniformly favorable and, undoubtedly, involves a complex interaction between host and viral elements.

Recent evidence has suggested that race and ethnicity may modify the likelihood of achieving a sustained virological response to interferon therapy (1–4). Reddy et al (1) reported that end-of-treatment and sustained virological responses were lower in black subjects compared to white subjects treated with consensus interferon at standard doses. Of 40 black subjects, only two (5%) had HCV RNA levels below detectable limits, as compared to 127 of 380 (33%) white subjects at the end of treatment. Sustained virological response occurred in only one of these black subjects (2%) while 12% (46/380) of white subjects remained negative for HCV RNA six months after treatment. Similar results were obtained when the data were limited to subjects with genotype 1, with the exception

Manuscript received February 16, 2002; revised manuscript received September 3, 2002; accepted September 4, 2002.

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Dr. Theodore is funded, in part, by NIH National Heart, Lung, and Blood Institute Minority Investigator Research Supplement (RO1-HL64817, PI: Michael W. Fried, MD).

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that sustained virological response was virtually identical for blacks and whites (3% vs 7%). In that study, median HCV RNA levels also decreased to a lesser extent in black subjects than in white subjects, and the mean change in HCV RNA levels during the course of therapy tended to be smaller in blacks (1). Thus, although race was correlated with decreases in HCV RNA levels during therapy in a logistic multiple regression model controlling for race, gender, baseline HCV RNA level, and genotype, the authors failed to demonstrate that race was associated with sustained virological response.

McHutchison et al performed a retrospective analysis of data from two large multicenter trials of interferon monotherapy versus interferon and ribavirin combination therapy for the treatment of chronic HCV (2). Subjects were treated with interferon monotherapy 3 MU thrice weekly or interferon and ribavirin combination therapy for either 24 or 48 weeks. Of 1744 subjects who were randomized and received treatment, only 53 (2.6%) subjects were black, but the data corroborate the findings of other investigators. No black participants responded to interferon monotherapy, while 88 of 675 (13%) white participants achieved a sustained virologic response. In addition, black subjects had significantly smaller reduction in HCV RNA levels during therapy compared to whites. This difference was most apparent in black subjects with genotype 1 infection who received interferon monotherapy.

To date, patients in these studies have all been treated with similar regimens of interferon (3 MU three times a week or 9 μg three times a week) alone or in combination with ribavirin. It is not known if a more aggressive interferon regimen would improve response rates among African Americans. We have previously reported that treatment with daily, high-dose interferon significantly improves initial and end-treatment response rates compared to standard doses of thrice weekly interferon. We conducted a retrospective analysis of our database, which included a high percentage of African Americans, to determine if an intensive regimen of daily, high dose interferon would improve the initial response rates to therapy for hepatitis C among African American patients compared to Caucasian patients.

MATERIALS AND METHODS

Details concerning study design and randomization have been reported previously (5). Briefly, subjects were eligible for participation if they had serologic evidence of hepatitis C, serum alanine aminotransferase activity >1.3 times the upper limit of normal, and detectable HCV RNA in serum by polymerase chain reaction. Subjects were evaluated for other causes of liver disease by appropriate serological screening and excluded from participation in the study if any such causes were identified. Subjects who had previously received interferon were excluded. Based upon earlier studies demonstrating that cirrhosis was an independent predictor of poor response to interferon, subjects with cirrhosis on a pretreatment liver biopsy were excluded. Likewise, subjects who tested positive for antibody to human immunodeficiency virus were excluded.

Between October 1995 and June 1997, one hundred four subjects were randomized to 24 weeks of therapy with interferon $-\alpha$-$_{2b}$ at either 5 MU *daily* or 3 MU three times a week with an additional 24-week period of posttreatment observation. Treatment groups were prospectively stratified by HCV genotype (1 or non-1) and pretreatment HCV RNA levels (< or >500,000 copies/ml) (Figure 1). Treatment was discontinued at week 12 of therapy for patients who remained HCV RNA positive, based upon recommendations concerning the predictability of sustained response at that time point for interferon monotherapy (6).

The study was conducted at eight referral centers in the United States. The protocol was approved by the human investigations committees at each participating institution. Written informed consent was obtained from each subject prior to enrolment in the study.

Nucleic Acid Analysis. Serum HCV RNA was analyzed using the Amplicor Monitor (version 1.0) assay (Roche Molecular Systems, Branchburg, New Jersey, USA). The linear dynamic range for this assay is 1×10^3–1×10^6 copies/ml. The lower limit of quantitation of HCV RNA was 2000 copies/ml. HCV RNA levels <1000 copies/ml were considered to be negative. HCV genotype was determined by the line probe assay (InnoLiPa, Innogenetics, Gent, Belgium).

Statistical Analysis. This analysis was limited to the subset of 74 African American and Caucasian subjects with HCV genotype 1. Stata (Stata Statistical Software: Release 6.0, 1999, Stata Corporation, College Station, Texas, USA) was used for all calculations. All data were analyzed by intention to treat. The χ^2 test was used to compare qualitative variables (or two-tailed Fisher's exact test when some expected values were <5) and Student's *t* test was used to compare means. Differences between groups for which $P \leq 0.05$ were considered statistically

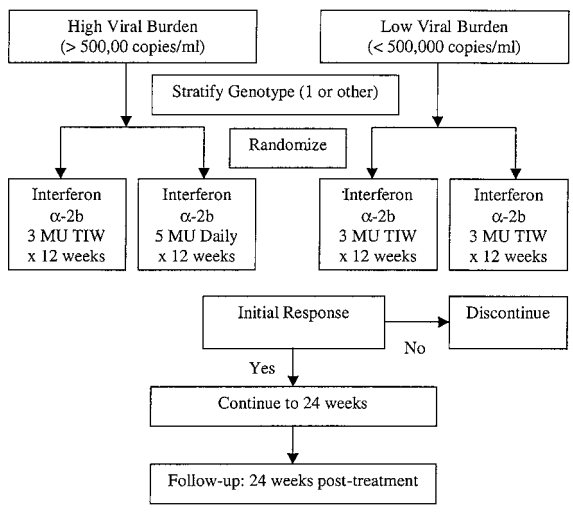

Fig 1. Study schema: Subjects were stratified by baseline HCV viral load and viral genotype.

significant. All 74 subjects were included in the survival analyses. The failure event was defined as having an HCV RNA that was below the limits of detection by week 12. Subjects were considered at risk once interferon therapy was initiated. The cutoff point for the analysis was the last date for which data were available for the subject. All subjects who had a virologic response had data available at their week 12 visit. Participants who were lost to follow-up were censored at the last date for which data were available. Subjects who had a virologic response but relapsed before week 12 were coded as if they had never experienced a response. The date of each subject visit was recorded and the unit of time for survival analysis was measured in days. All explanatory variables were measured before the time origin. There were no missing values among the explanatory variables.

RESULTS

Of the 104 study participants from the parent study, African Americans numbered 21 (20%) and Caucasians 78 (75%). Genotype 1 was significantly more common among African Americans than Caucasians [20/21 (95%) vs 54/78 (69%); $P < 0.02$]. The baseline demographic characteristics of the subjects in the retrospective cohort are presented in Table 1. There were no statistically significant differences between African American subjects with respect to mean age, gender distribution, and likelihood of receiving treatment with interferon 5 MU daily. African American subjects in the parent study were more likely to be classified in the high-viral-burden group than Caucasian subjects, but among African American and Caucasian participants with genotype 1, mean levels of HCV RNA were similar at baseline between the two racial groups (1.37×10^6 vs 6.38×10^6, $P =$ NS).

The response rates at week 12 among African Americans or Caucasians for two IFN regimens are shown in Table 2. Thirty percent (9/30) of non-Hispanic white patients had an initial response (defined as undetectable HCV RNA at week 12 of treatment) when treated with the standard interferon regimen of 3 MU three times a week compared to 8% (1/12) of African Americans ($P =$ NS). However, with the more intensive, daily interferon regimen, white subjects were more likely than black subjects to show an initial response (75% vs 25%, $P = 0.03$). Furthermore, Caucasian subjects who received the more intensive interferon regimen had a significantly better initial response to therapy than did Caucasians treated with the standard interferon dosing (75% vs 30%, $P < 0.002$). In contrast, a more intensive regimen did not improve the initial response among African American subjects (25% vs 8%, $P =$ NS).

TABLE 1. DEMOGRAPHIC AND VIROLOGIC CHARACTERISTICS AT BASELINE

	African American (N = 20)	*Caucasian (N = 54)*	P
Age (yr)	45 ± 9	42 ± 7	NS
Male gender	12 (60%)	35 (65%)	NS
Interferon 5 MU	8 (40%)	24 (44%)	NS

TABLE 2. INITIAL RESPONSE TO TWO INTERFERON DOSAGE REGIMENS IN AFRICAN AMERICAN OR NON-HISPANIC WHITE PATIENTS WITH GENOTYPE 1

	Initial Response [N (%)]		
Interferon dose group	*African American*	*Non-Hispanic white*	P
5 MU daily	2/8 (25%)a	18/24 (75%)b	0.03 (a vs b)
3 MU (3 ×/week)	1/12 (8%)c	9/30 (30%)d	NS (c vs d)
P	NS (a vs c)	<0.002 (b vs d)	

The proportion of subjects with an undetectable HCV viral load was evaluated at each study visit. The percent of subjects with a viral load below the limits of detection was stratified by race and dose of interferon and plotted against time (Figure 2). At each visit beyond the week 2 visit, the proportion of Caucasian subjects with an undetectable HCV RNA was greater than comparable African American subjects, although the difference did not reach statistical significance until the week 12 visit, and only in the intensive treatment groups ($P = 0.03$).

The changes in geometric mean levels of HCV RNA at various times among African Americans and Caucasians treated with either interferon regimen are shown in Figure 3. The initial response to therapy was significantly greater for Caucasians treated with intensive interferon compared to either African American group or for white subjects treated with standard interferon.

To determine whether or not time to becoming and staying HCV RNA negative was different between African

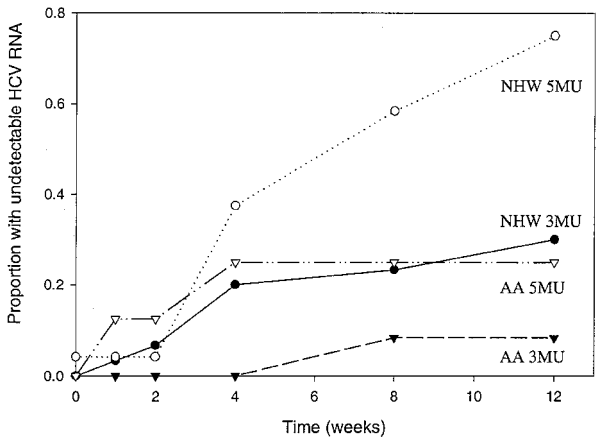

Fig 2. Proportion of African American and non-Hispanic white subjects with an undetectable HCV RNA during the initial 12 weeks of treatment.

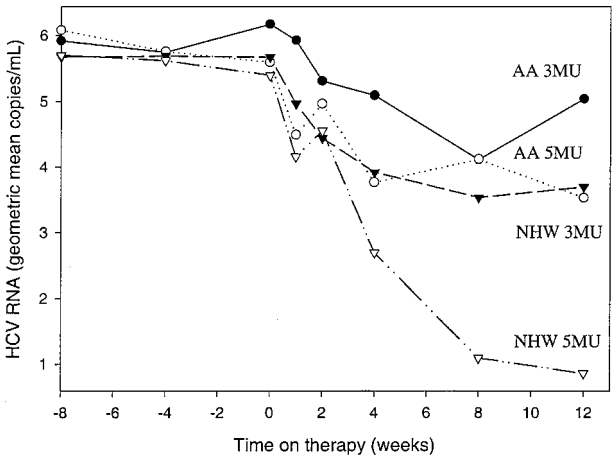

Fig 3. Serial changes of HCV RNA during the initial 12 weeks of treatment for each race/dose group. The *y* axis shows the $\log_{10}$ geometric mean copies per milliliter for HCV RNA.

Americans and Caucasians, we carried out a survival analysis using all 74 subjects. There were 30 failures (those reaching the end point of negative HCV RNA in serum). The total time at risk was 4930 days. Twenty-five percent of subjects had achieved a virologic response by 56 days. The Nelson-Aalen cumulative hazard estimates are graphically presented for each race and dose combination (Figure 4). African Americans receiving standard doses of interferon appeared to have the worst response, while Caucasian subjects receiving high dose intensive therapy seemed to respond the best. Caucasians allocated to the standard interferon regimen and African Americans on the intensive daily regimen had an intermediate response. We compared the survival probabilities of the different race

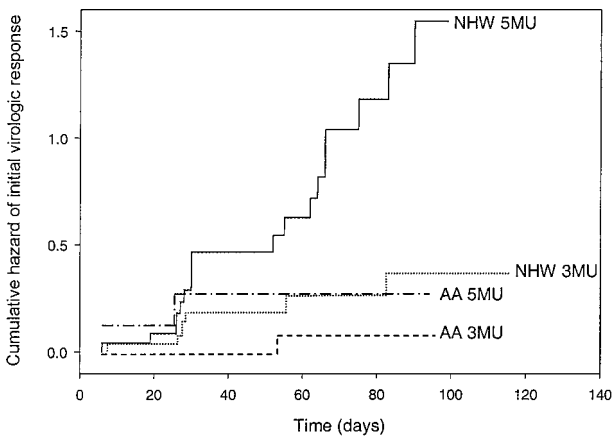

Fig 4. Nelson-Aalen cumulative hazard estimates of the likelihood of achieving an undetectable HCV RNA by week 12 of therapy. NHW = non-Hispanic white; AA = African American; 3MU = Interferon 3MU three times a week; 5MU = interferon 5MU daily.

and dose combinations and found that they were statistically different (logrank, $P < 0.001$).

Univariate analysis of explanatory variables was conducted to explore correlates of virologic response by week 12. We examined five binary variables (race, interferon dose, past alcohol use, gender, pretreatment viral burden) using the logrank test. Viral burden was dichotomized *a priori* in the parent study. Subjects with HCV RNA levels <500,000 copies/ml were considered to have a low viral burden (3). All other subjects were classified in the high-viral-load group. Age was analyzed as a continuous variable using univariate Cox regression. We also analyzed site of treatment as a categorical variable using the generalized Wilcoxon test of Breslow. Caucasians were more likely to respond to interferon than African American subjects ($P < 0.03$). Likewise, subjects allocated to the more intensive interferon regimen were more likely to have an initial virologic response than subjects treated with standard thrice-weekly interferon injections ($P < 0.001$). We also evaluated dose by race. The more intensive regimen was more effective than the standard regimen among Caucasian subjects ($P < 0.01$) but not among African American subjects, although there were fewer subjects in the African American stratum. Past alcohol use and gender were not associated with virologic response by week 12. Low viral burden, on the other hand, was a predictor of response ($P < 0.01$). Age was not found to be a significant correlate of response. Treatment sites did vary with respect to initial virologic outcome, but this did not reach statistical significance ($P = 0.06$).

We subsequently specified a multivariate Cox proportional hazards model using all the covariates evaluated in our univariate analysis. We included these variables because they have been reported in the medical literature as potential predictors of response. Site was included based on the results of the univariate analysis. The model was reduced by removing the most disputable covariates (treatment site, past alcohol consumption, age) and rerunning the model with the remaining variables. The likelihood ratio test confirmed that the variables eliminated did not contribute significantly to the model. The proportional hazards assumption was evaluated graphically and found to be appropriate. The exponentiated final model is presented in Table 3. The model shows that the likelihood of achieving and maintaining a virologic response by week 12 of therapy is twice as great ($P < 0.01$) for subjects receiving intensive interferon therapy compared to subjects on standard interferon, controlling for the effects of race, gender, and pretreatment viral burden. While African American subjects and men appeared to fare worse than Caucasians and women, respectively, these findings were not statistically significant. Controlling for the contributions of race,

TABLE 3. RELATIVE HAZARDS (RH) OF INITIAL VIROLOGIC RESPONSE AND 95% CONFIDENCE INTERVAL FROM COX PROPORTIONAL HAZARDS MODEL

Model covariate	*RH (95% CI)*	P
Race	0.333 (0.099–1.123)	0.076
Dose	1.998 (1.352–2.954)	0.001
Gender	0.496 (0.240–1.025)	0.058
Pretreatment viral load	2.726 (1.293–5.750)	0.008

dose, and gender, the likelihood of a virologic response was almost three times greater for subjects with a low pretreatment viral load compared to participants with a high HCV RNA ($P < 0.01$).

A sustained virologic response (measured 24 weeks after cessation of therapy) was seen in only one African American patient (11%) treated with the high-dose regimen compared to none in the standard-dose regimen. In Caucasian patients treated with high-dose interferon, 10% had a sustained virologic response compared to 4% of those on the standard regimen. These differences were not statistically significant.

Adverse events have been previously described in detail (5). Interferon was well tolerated in both the high-dose regimen and the standard-dose regimen. Individual adverse events occurred too infrequently to allow for meaningful comparisons, but overall there was no difference in adverse events between African American and Caucasian participants (data not shown).

DISCUSSION

Recent secondary analyses of large databases derived from clinical trials of various antiviral agents have suggested that there may be racial disparities in response to therapy for hepatitis C. Sustained response rates in African American patients appear to be significantly lower than in Caucasian patients treated with the same regimens. However, these trials were not designed specifically to test the hypothesis that race was a factor in response to therapy, and very few African Americans have been included as study subjects in all of the published registration trials of antiviral agents for this disease. While the current study also was not prospectively designed for this, the trial contained a high proportion of African American subjects, which closely mirrors the proportion of individuals with hepatitis C who are African American (3).

Previously published reports that evaluated race and response to interferon therapy have all utilized standard interferon dosing regimens (3MU three times a week or 9 μg three times a week) (4, 7). From studies of viral kinetics after a single injection of interferon in patients with genotype 1, higher doses of interferon (5–15 MU) did result in more rapid, greater decreases of HCV RNA when compared to a standard dose of interferon (8–10). Thus, it is possible that patients with characteristics of interferon resistance might respond better to a more aggressive interferon regimen (11). Kinzie et al (12) used a dose escalation strategy in which subjects without an early virological response after 12 weeks of interferon were either dose-escalated to 5 MU three times a week or continued at 3 MU three times a week. They found that African Americans were less likely to achieve an end of treatment response compared to Caucasians. The more intensive treatment used in the current study, which prospectively randomized patients to a daily, high dose regimen or a standard regimen, provided an opportunity to test the relationship between dose and response across racial groups.

The results of the current study corroborate reports that African American patients with hepatitis C were more likely to be infected with treatment resistant genotype 1 than were non-Hispanic whites (1, 2, 13, 14). Our analysis of antiviral response was limited to only those patients, African American or white, that had HCV genotype 1, thus eliminating this potentially confounding variable.

The results presented here demonstrate that an intensive regimen of daily high dose interferon is superior to standard interferon with respect to the proportion of subjects who have an initial virologic response and with respect to time to achieving such a virologic response. Of note, we were able to show a dose response to interferon among Caucasian participants, but no such effect was demonstrable among African American subjects.

Because of its retrospective nature, it is worth noting some limitations of the current study. Although we did prospectively stratify subjects on the basis of low or high-viralburden, we did not stratify by race. This may explain why African American subjects were more likely than Caucasian subjects to be classified in the high-viral-burden group. However, when we limited the data to the subgroup of African American and Caucasian subjects with genotype 1, the mean HCV viral load at baseline was similar. We did not specifically inquire about adherence to medications, nor did we include body mass index, baseline weight, or pretreatment liver histology in our study. In other published investigations, the contribution of these factors to interferon response rates has been inconclusive. Thus, the extent to which these potential confounders help to explain the differences observed in this study between African American and Caucasian patients with genotype 1 cannot be determined.

Another limitation of the study is based on the classification of subjects by race (15–18). Because race is primarily a social construct, it is possible that observed

differences between African American and Caucasian subjects are behavioral and environmental in nature, rather than being intrinsic to being African American or Caucasian. Race in this sense may merely be a surrogate for something that we have failed to measure or adequately control for in our analyses. Although the proportion of African American subjects in this study is reflective of the percentage of HCV patients who are African Americans, the total sample size may have been inadequately powered to show small but clinically significant differences.

Despite these limitations, this is the first study to examine whether or not a significantly more intensive interferon regimen would improve response rates in African Americans. Treatment with a daily high-dose regimen improved the initial response for Caucasians but had little effect on response in African Americans. Thus, it appears that increasing the dose and frequency of interferon does not overcome the antiviral resistance in African Americans. This may be of immediate relevance because the pharmacokinetics of pegylated interferons mirrors in some respects the daily high-dose interferon regimen used in the current study. The data suggest that African American patients may have resistance to interferon that may be incompletely overcome by the use of pegylated interferons. Indeed, preliminary reports suggest that the virological response in African Americans is improved with pegylated interferon monotherapy but remains less than that in Caucasian subjects (19, 20). Future studies should carefully and prospectively evaluate the effect of race on response to antiviral treatment for chronic HCV and investigate potential mechanisms for diminished response to interferon.

ACKNOWLEDGMENTS

We thank Drs. William C Miller and Marcia Hobbs for their review of the manuscript.

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