Favorable Prognosis of Chronic Hepatitis C After Interferon Therapy by Long-Term Cohort Study

Fumio Imazeki, Osamu Yokosuka, Kenichi Fukai, and Hiromitsu Saisho

The prognosis of patients with chronic hepatitis C after interferon (IFN) therapy is still poorly defined. The present study evaluated the effect of IFN therapy on survival in a cohort of such patients. The study included 459 patients with biopsy-proven C-viral chronic liver disease who were followed for 8.2 ± 2.9 years (range, 7-183 months). Survival status was examined by medical records or direct questionnaires. Fifteen (14%) of 104 IFN-untreated patients and 33 (9%) of 355 patients treated with IFN died during follow-up. Among the treated patients, 4 (3%) of 116 with sustained virologic response and 29 (12%) of 239 without sustained virologic response died. Liver-related death was shown in 32 (67%) patients, and hepatocellular carcinoma (HCC) caused 25 (52%) of the 48 deaths. Multivariate Cox proportional regression analysis revealed that IFN treatment decreased the risk ratio for overall death to 0.521 (confidence interval [CI]: 0.263-1.034) and for liver-related death to 0.208 (CI: 0.088-0.495) compared with untreated patients, and sustained virologic response showed a decrease in the risk ratio for overall death to 0.219 (CI: 0.068-0.710) and for liver-related death to 0.030 (CI: 0.003-0.267). IFN treatment showed no association with liver-unrelated death. Furthermore, the standardized mortality ratios for all causes of death and liver-related death were reduced in IFN-treated patients compared with untreated patients (1.4 vs. 2.0 for total death and 7.9 vs. 19.7 for liver-related death). In conclusion, the present data suggest that IFN therapy has a long-term clinical benefit for patients with chronic hepatitis C patients by reducing liver-related death, especially in patients with sustained virologic response. (HEPATOLOGY 2003;38:493-502.)

See Editorial on Page 292

Hepatitis C virus (HCV) infection has been shown to be closely associated with chronic hepatitis, leading to cirrhosis and hepatocellular carcinoma throughout the world.[1,2] In Japan, more than 30,000 people die of liver neoplasms annually. Most of these deaths are associated with hepatocellular carcinoma (HCC), and around 80% are associated with HCV infection.[3] Additionally, about 10,000 people die of cirrhosis or chronic liver disease excluding alcoholic liver disease. Thus, HCV-related chronic liver disease is one of the major disorders affecting the national health of Japan.

Interferon (IFN) is effective in eliminating HCV and reducing the alanine aminotransferase (ALT) level in some patients with chronic hepatitis C,[4,5] and improvement in liver histologic findings have also been noted in these patients.[6,7] IFN monotherapy achieves sustained virologic response in 20% to 40% of chronic hepatitis C, with the response rate differing somewhat according to various factors such as viral load, viral genotype, stage of liver fibrosis, and the total dose of IFN.[8-10] Several recent studies showed that IFN treatment reduced the risk for development of HCC in comparison with untreated patients and especially in those who responded to the therapy.[11-13] However, it is still a matter of controversy whether the effect of IFN treatment on C-viral cirrhotic patients is beneficial or not.[14-18]

Several studies concerning the prognosis of C-viral cirrhosis patients after IFN therapy have been reported, but their results were also controversial.[19-22] Furthermore, there have been only a few reports concerning those with chronic hepatitis C.[23-25] In the current investigation, we conducted a retrospective cohort study to examine the effect of IFN therapy on the long-term prognosis of pa-

Abbreviations: HCV, hepatitis C virus; HCC, hepatocellular carcinoma; IFN, interferon; ALT, alanine aminotransferase; AST, aspartate aminotransferase; SMR, standardized mortality ratio; CI, confidence interval.

From the Department of Medicine and Clinical Oncology, Graduate School of Medicine, Chiba University, Chiba, Japan.

Received October 3, 2002; accepted May 15, 2003.

Address reprint requests to: Fumio Imazeki, 1-8-1 Inohana, Chuou-ku, Chiba City, Chiba, Japan 260-8670. E-mail: imazeki@med.m.chiba-u.ac.jp; fax: (81) 43-226-2088.

0270-9139/03/3802-0027$30.00/0
doi:10.1053/jhep.2003.50329

tients with C-viral chronic liver disease in comparison with untreated patients.

Patients and Methods

Patients. Four hundred seventy-two consecutive patients with C-viral chronic liver disease who underwent liver biopsy at the First Department of Medicine, Chiba University Hospital, between January 1986 and December 1998, were enrolled in this study. Of these, 13 patients were excluded: Nine of them were referred to other hospitals within 6 months after liver biopsy and dropped out, and the other 4 were excluded because HCC was detected within 6 months after liver biopsy and the possibility of the presence of hepatocellular carcinoma at the time of liver biopsy could not be ruled out. The study group consisted of 280 men and 179 women with a mean age of 50.1 ± 12.0 years. The absence of HCC was ascertained at enrollment by abdominal ultrasonography, computed tomography, or magnetic resonance imaging. Patients with chronic hepatitis B, autoimmune hepatitis, primary biliary cirrhosis, hemochromatosis, Wilson's disease, and alcoholic liver disease were excluded from this study.

Liver biopsy specimens were examined by 2 independent liver pathology specialists (F.I. and O.Y.), blinded to the clinical and virologic results according to the criteria of Desmet et al.,[26] with the staging of fibrosis being defined as F0 (no fibrosis), F1 (mild fibrosis), F2 (moderate fibrosis), F3 (severe fibrosis), and F4 (cirrhosis).

Laboratory and Imaging Examination. All patients in this study were positive for HCV-Ab as determined by second-generation enzyme-linked immunosorbent assay. The serum HCV load of the patients was quantitatively determined at the time of liver biopsy using various commercial and in-house assays such as competitive reverse-transcription[27] and branched-DNA probe assay[28] prospectively or retrospectively. However, because it is difficult to correlate the results of different assays, we adopted HCV core protein assay[29] for the quantification of serum HCV load at the time of liver biopsy using stored sera of all the patients. The sera used for HCV core protein assay were stored at −20°C, and the duration of storage before assay was 6.1 ± 2.4 and 5.9 ± 2.7 years in treated and untreated patients, respectively. HCV RNA genotype was determined by serologic grouping of serum antibody,[30] assuming that genotypes 1a and 1b correspond to group 1 and genotypes 2a and 2b correspond to group 2.

Abdominal ultrasonography was performed every 3 to 6 months to detect HCC, and space-occupying lesions detected were further examined by computed tomography, magnetic resonance imaging, hepatic arteriography, and ultrasound-guided tumor biopsy if necessary.

IFN Treatment. IFN was administered to 355 patients, and 104 patients did not receive IFN treatment. IFN therapy was initiated within 1 year after liver biopsy. Eighty-four percent of treated patients received IFN-α, 12% IFN-β, and 4% both. The median total dose and duration of IFN administration was 468 megaunits (72-2,030 megaunits) and 167 days (6-560 days). Once IFN therapy was started, a patient was included in the IFN treatment group even if therapy was discontinued because of adverse effects or other reasons. The 104 patients not receiving IFN chose this course on the basis of concerns about adverse effects (32%), not having health insurance coverage (27%), normal or nearly normal ALT (13%), lack of sufficient time to undergo therapy (10%), physician's decision based on the presence of depression (4%), cardiopulmonary disease (3%), other concerns (5%), and no known specific reason (6%).

Virologic criteria were used to define response to IFN therapy. A virologic sustained response was defined as HCV RNA negativity, determined by reverse transcription-polymerase chain reaction, more than 6 months after termination of IFN therapy. HCV RNA positivity at the same time point was considered a nonsustained response.

No patients in our series died during IFN therapy, and only 1 patient died within 6 months after its termination and was classified as nonresponder because of positive serum HCV RNA at the end of treatment.

Survival Status and Cause of Death. The follow-up period was defined as the duration between the date of liver biopsy and either the date of death or the latest date of confirmed survival. Survival status was confirmed by medical records or direct survey conducted by telephone. Three hundred fourteen of the patients visited our hospital periodically, and their survival status could be confirmed by medical records. The survival status of 95 of 145 patients who stopped visiting our hospital was confirmed by direct communication via telephone, but the remaining 50 patients were lost to follow-up and were included in the analysis as alive up to the last available point of contact. Hence, 409 patients (89%) in the cohort had their survival status confirmed at the last examination in February 2002. The cause of death was investigated from death certificates in 44 of 48 (92%) patients and that of the other 4 patients, whose cause of death was liver failure, subarachnoidal hemorrhage, rupture of abdominal arterial aneurysm, and traumatic brain damage, was obtained from telephone interviews with their families.

Cause of death was divided into liver related and unrelated. The former included HCC, cholangiocellular carcinoma, liver failure, and esophagogastrial variceal

Table 1. Baseline Characteristics of Patients in the Cohort Study

Characteristic	Interferon Treated	Untreated	*P* Value
Patients, n	355	104	
Sex (male/female), n	228/127 (64%/36%)	52/52 (50%/50%)	.0117
Age (y), mean ± SD	49.2 ± 11.9	53.1 ± 11.4	.0033
Fibrosis stage			
F0/F1/F2/F3/F4	14/185/61/49/46 (4%/52%/17%/14%/13%)	5/53/15/9/22 (5%/51%/14%/9%/21%)	.2090
AST level (IU/L), mean ± SD	92 ± 66	68 ± 40	.0003
ALT level (IU/L), mean ± SD	141 ± 107	87 ± 61	<.0001
Albumin level (g/dL), mean ± SD	4.3 ± 0.3	4.2 ± 0.4	.0091
Platelet count ($\times 10^9$/L), mean ± SD	171 ± 60	166 ± 73	.4180
HCV core protein (pg/mL), mean ± SD	171 ± 212	230 ± 298	.0314
HCV genotype (1/not 1), n	246/86 (74%/26%)	68/25 (73%/27%)	.8939
Alcohol consumption ≥20 g/d, n	64 (18%)	18 (17%)	>.9999
Body mass index (kg/m^2)	23.0 ± 3.0	23.1 ± 3.1	.9398
Mode of infection (%)			
Blood transfusion	159 (45)	51 (49)	.5757
Drug addiction	37 (10)	12 (12)	.7206
Tattoo	8 (2)	3 (3)	.7775
Unspecified	151 (43)	38 (36)	.1637
Duration of the disease (y), mean ± SD	23.3 ± 13.6	24.5 ± 11.4	.5122
Comorbidities (%)			
Diabetes	25 (7)	11 (11)	.2995
Hypertension	38 (11)	9 (9)	.5861
Fatty liver	23 (7)	3 (3)	.2276
Cardiopulmonary disease	13 (4)	4 (4)	>.9999
Autoimmune disease	7 (2)	2 (2)	>.9999
Psychotic disease	2 (0.6)	3 (3)	.0793
Others	5 (1)	4 (4)	.9685

bleeding, and the latter included extrahepatic malignancy, heart diseases, cerebrovascular accidents, and others (Table 10).

Statistical Analysis. Student's *t* test and Fisher exact test were used to analyze quantitative and qualitative data, respectively. Cox proportional regression analysis was performed to estimate rate ratios for the effect of IFN therapy for survival. Potential risk factors assessed for survival included the following variables: age (≥60 years, 50-59 years vs. ≤49 years); sex; stage of liver fibrosis (F2, F3, F4 vs. F0/1); IFN treatment; IFN efficacy; baseline aspartate aminotransferase (AST) level (≥80 IU/L vs. <80 IU/L); baseline ALT level (≥80 IU/L vs. <80 IU/L); baseline albumin level (≥4.2 g/dL vs. <4.2 g/dL); baseline platelet count (≥13 × 10^9/L vs. <13 × 10^9/L); HCV load (HCV core protein ≥100 pg/mL vs. <100 pg/mL); HCV serotype (1 vs. not 1); alcohol consumption (≥20 g/d vs. <20 g/d); duration of the disease (≥20 years vs. <20 years); body mass index (≥25 vs. <25); and comorbidities such as diabetes mellitus, hypertension, fatty liver, and cardiopulmonary disease. Variables statistically significant by univariate Cox proportional regression analysis were further studied by multivariate analysis. Cumulative occurrence curves of survival were determined by the Kaplan-Meier method, and the differences between groups were assessed by the log-rank test. *P* value less than 0.05 was considered to indicate statistical significance.

Standardized mortality ratios (SMR) were used to compare overall cause and specific cause of mortality of the total cohort and of subgroups with a general population matched for age and sex. Death rates determined from published sources in 1990, 1995, and 2000[31] were used to calculate the expected number of deaths according to person-years of observation in various age (5 year) ranges and by gender. A *P* value less than .05 was considered significant for the ratio of an observed value of a Poisson variable to its expectation.

Results

Patient Characteristics. The demographic and clinical features of patients at enrollment are summarized according to IFN treatment in Table 1. Characteristics of sex, age, AST level, ALT level, albumin level, and HCV core protein level were statistically different between IFN-treated and -untreated groups (Table 1). The proportions of patients with comorbidities such as diabetes, hypertension, fatty liver, cardiopulmonary diseases, autoimmune disease, psychotic disease, and others were almost identical between the 2 groups (Table 1). The presumed duration of the disease, determined from the time of blood

transfusion, the start of drug addiction, or the time of tattoo until the time of liver biopsy, was 23.3 ± 13.6 years and 24.5 ± 11.4 years in IFN-treated and -untreated patients, respectively ($P = .5122$) (Table 1). Regarding the laboratory data and clinical stage of cirrhotic patients in the IFN-treated and -untreated groups, the albumin level was 4.0 ± 0.4 vs. 3.9 ± 0.5 ($P = .3431$), total bilirubin 1.0 ± 0.4 vs. 0.9 ± 0.3 ($P = .3765$), Child-Pugh score 5.2 ± 0.5 vs. 5.3 ± 0.6 ($P = .4987$), and presence of esophageal varices 5 of 46 (11%) vs. 6 of 22 (27%) ($P = .1556$) in IFN-treated and -untreated patients, respectively. None of the cirrhotic patients had ascites or hepatic encephalopathy at entry. The follow-up period was 8.2 ± 2.9 years (mean ± SD; range, 7-183 months; median, 101 months) from the date of liver biopsy to the date of final medical examination or to the date of ascertaining their survival status by telephone: 8.3 ± 2.9 years in IFN-treated and 7.8 ± 2.9 years in IFN-untreated patients ($P = .1125$).

Response to IFN therapy was determined in 355 patients, with a virologic sustained response being achieved in 116 (33%), and the remaining 239 (67%) patients showing a nonsustained response (Table 2).

Mortality Rate and Underlying Causes of Death. Forty-eight (10%) of 459 patients died during the follow-up period. They consisted of 15 (14%) of 104 untreated and 33 (9%) of 355 IFN-treated and 4 (3%) of 116 virologic sustained responders and 29 (12%) of 239 nonsustained responders (Table 2). The annual mortality rate calculated by the person-years method was 1.8% in untreated patients and 1.1% in IFN-treated patients, the latter consisting of 0.4% of virologic sustained responders and 1.3% of nonsustained responders. Death caused by liver-related disease was seen in 32 (67%) cases of overall death, 19 (58%) of 33 IFN-treated and, in contrast, in 13 (87%) of 15 untreated patients.

Table 2. Mortality Rate and Underlying Causes of Death in Patients With HCV-Related Chronic Liver Disease

	Untreated	IFN Treated	SVR	NSR
Cases	104	355	116	239
No. of deaths (%)	15 (14)	33 (9)	4 (3)	29 (12)
Observation period (y), mean ± SD	7.8 ± 2.9	8.3 ± 2.9	8.3 ± 3.2	8.3 ± 2.7
Annual mortality rate, %	1.8	1.1	0.4	1.3
Cause of death (%)				
HCC	11 (72)	14 (43)	1 (25)	13 (46)
Hepatic failure	1 (7)	4 (12)	0	4 (14)
Variceal bleeding	0	1 (3)	0	1 (3)
Other cancers	1 (7)	4 (12)	1 (25)	3 (10)
Cerebrovascular diseases	1 (7)	4 (12)	1 (25)	3 (10)
Others	1 (7)	6 (18)	1 (25)	5 (17)

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder.

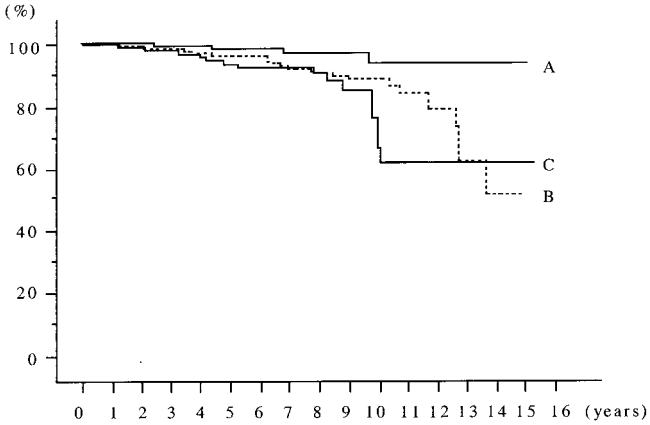

Fig. 1. Cumulative survival curves of virologic sustained responders (A), nonsustained responders (B), and IFN-untreated patients (C). *P* value was less than .05 by log-rank test between virologic sustained responders (n = 116) and nonresponders (n = 239) or IFN-untreated patients (n = 104).

HCC developed in 63 patients during follow-up, and it was the most common cause of death in the cohort. Twenty-five patients (52%) died of HCC, 11 (72%) of 15 untreated and 14 (43%) of 33 IFN-treated (Table 2). Only 1 of the virologic sustained responders died of HCC, and the other 3 died of liver-unrelated diseases (Table 2).

The mean age at the time of death of the 48 patients was 64.1 ± 7.6 years. There was no statistical difference among untreated patients, sustained virologic responders, and nonsustained responders (63.6 ± 7.4 vs. 64.3 ± 6.8 vs. 64.3 ± 8.1, respectively). There was also no difference between patients who died of liver-related diseases and of liver-unrelated diseases (64.4 ± 7.3 vs. 63.5 ± 8.6, respectively, $P = .7031$).

Cumulative survival curves for overall death between virologic sustained responders and nonsustained responders or IFN-untreated patients and statistical significance are shown in Fig. 1.

IFN Effect on the Risk of Overall Death by Multivariate Analysis. The potential risk factors affecting overall death were analyzed by univariate Cox proportional hazards regression (Table 3). Statistically significant variables were further studied by multivariate analysis (Table 4). Age, sex, fibrotic stage, IFN treatment, and IFN efficacy were shown to be associated with risk for overall death. IFN treatment showed a decrease in the risk ratio for overall death to 0.521 (confidence interval [CI]: 0.263-1.034) compared with untreated patients to a marginally significant degree ($P = .0622$), but, when the IFN treatment group was divided into virologic sustained and nonsustained responders, the former showed a decrease in the risk ratio to 0.219 (CI: 0.068-0.710) (Table 4). The

Table 3. Risk for Overall Death by Univariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	*P* Value
Age			.0002
<49 y	1.0		
50-59 y	8.867	3.096-25.395	<.0001
≥60 y	8.100	2.720-24.122	.0002
Male (vs. female)	2.161	1.120-4.168	.0215
Fibrotic stage			<.0001
F0/1	1.0		
F2	5.875	2.171-15.904	.0005
F3	9.068	3.402-24.171	<.0001
F4	17.304	6.850-43.713	<.0001
Interferon treatment			.0194
Untreated	1.0		
Treated	0.585	0.317-1.080	.0862
SVR	0.207	0.069-0.625	.0052
NSR	0.782	0.418-1.464	.4419
AST ≥ 80 IU/L (vs. <80)	3.282	1.761-6.118	.0002
ALT ≥ 80 IU/L (vs. <80)	2.533	1.226-5.235	.0121
Albumin ≥ 4.2 g/dL (vs. <4.2)	0.397	0.224-0.702	.0015
Platelet ≥ 13×10^9/L (vs. <13×10^9)	0.224	0.125-0.399	<.0001
HCV core protein ≥ 100 pg/mL (vs. <100)	1.452	0.802-2.627	.2179
Genotype 1 (vs. not 1)	1.799	0.839-3.857	.1315
Alcohol consumption <20 g/d (vs. ≥20 g/d)	0.478	0.256-0.893	.0206
Duration of the disease ≥20 y (vs. <20 y)	2.775	1.105-6.973	.0299
BMI ≥ 25.0 (vs. <25.0)	1.844	0.925-3.676	.0823
Comorbidities (vs. no comorbidities)	1.703	0.948-3.060	.0749
Diabetes (vs. no diabetes)	1.583	0.670-3.736	.2948
Hypertension (vs. no hypertension)	2.031	0.981-4.205	.0564
Fatty liver (vs. no fatty liver)	0.920	0.222-3.819	.9091
Cardiopulmonary disease (vs. no disease)	1.262	0.391-4.076	.6974

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder; BMI, body mass index (kg/m^2).

effect of IFN treatment on the risk of overall death of cirrhotic patients was also assessed by multivariate Cox proportional hazards regression adjusted for sex and age, with the risk being reduced to 0.348 (CI: 0.134-0.905).

IFN Effect on the Risk of Liver-Related and Unrelated Death by Multivariate Analysis. The potential risk factors affecting liver-related death were analyzed by univariate Cox proportional hazards regression (Table 5). Statistically significant variables were further studied by multivariate analysis (Table 6). Fibrotic stage, IFN treatment, IFN efficacy, AST level, and alcohol consumption were shown to be associated with risk for liver-related death. IFN treatment showed a decrease in the risk ratio for liver-related death to 0.208 (CI: 0.088-0.495) compared with untreated patients. Both virologic sustained and nonsustained responders after IFN therapy showed a decrease in risk ratio, the former to 0.030 (CI: 0.003-0.267) and the latter to 0.257 (CI: 0.108-0.609) (Table 6). The effect of IFN treatment on the risk of liver-related death of cirrhotic patients was also assessed by this method adjusted for sex and age, and there was a reduction in the risk to 0.181 (CI: 0.050-0.663).

The potential risk factors affecting liver-unrelated death were analyzed by univariate Cox proportional hazards regression (Table 7). Overall comorbidity was associated with risk for liver-unrelated death, although each of the comorbidities such as diabetes, hypertension, fatty liver, or cardiopulmonary diseases was not.

Statistically significant variables were further studied by multivariate analysis (Table 8). Sex and fibrotic stage were shown to be associated with risk for liver-unrelated death and overall comorbidity was not. Neither IFN treatment nor IFN efficacy affected liver-unrelated death. Cirrhosis was shown to be an independent risk factor for liver-unrelated death as well as liver-related death. Liver-unrelated diseases caused death in 8 of 19 (42%) cirrhotic patients and in 8 of 29 (28%) noncirrhotic patients.

SMR in the Cohort. Survival in the total cohort of patients with C-viral chronic liver disease was reduced when compared with survival expected for a matched general population. SMR for all causes was 1.6, statistically significant ($P < .01$) (Table 9). SMR for liver neoplasms and cirrhosis/chronic liver disease was 12.6 and 5.9, respectively, in patients with HCV infection ($P < .01$), whereas SMR for extrahepatic neoplasms was reduced to 0.4 ($P < .05$), and SMR for cerebrovascular accident was relatively increased without statistical significance. None

Table 4. Risk for Overall Death by Multivariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	*P* Value
Age			.0411
<49 y	1.0		
50-59 y	4.081	1.358-12.261	.0122
≥60 y	3.775	1.200-11.879	.0231
Male (vs. female)	2.667	1.291-5.510	.0081
Fibrotic stage			.0013
F0/1	1.0		
F2	2.617	0.886-7.726	.0815
F3	4.063	1.374-12.019	.0113
F4	7.165	2.555-20.091	.0002
Interferon treatment			.0397
Untreated	1.0		
Treated	0.521	0.263-1.034	.0622
SVR	0.219	0.068-0.710	.0114
NSR	0.633	0.317-1.262	.1937
AST ≥ 80 IU/L (vs. <80)	2.377	0.961-5.881	.0610
ALT ≥ 80 IU/L (vs. <80)	0.925	0.325-2.635	.8837
Albumin ≥ 4.2 g/dL (vs. <4.2)	0.792	0.427-1.467	.4579
Platelet ≥ 13×10^9/L (vs. <13×10^9)	0.639	0.329-1.239	.1846
Alcohol consumption <20 g/d (vs. ≥20 g/d)	0.724	0.353-1.484	.3772
Duration of the disease ≥20 y (vs. <20 y)	0.673	0.238-1.902	.4549

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder.

Table 5. Risk for Liver-Related Death by Univariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	*P* Value
Age			.0021
<49 y	1.0		
50-59 y	13.566	3.163-58.184	.0004
≥60 y	9.990	2.183-45.709	.0030
Male (vs. female)	1.598	0.753-3.392	.2221
Fibrotic stage			<.0001
F0/1	1.0		
F2	14.437	3.115-66.907	.0006
F3	22.841	5.002-104.293	<.0001
F4	32.360	7.107-147.355	<.0001
Interferon treatment			.0120
Untreated	1.0		
Treated	0.376	0.185-0.764	.0069
SVR	0.056	0.007-0.432	.0056
NSR	0.550	0.268-1.129	.1032
AST ≥80 IU/L (vs. <80)	4.806	2.078-11.119	.0002
ALT ≥80 IU/L (vs. <80)	2.485	1.022-6.047	.0447
Albumin ≥4.2 g/dL (vs. <4.2)	0.468	0.233-0.942	.0334
Platelet ≥13 × 10^9/L (vs. <13 × 10^9)	0.176	0.085-0.366	<.0001
HCV core protein ≥100 pg/mL (vs. <100)	1.756	0.845-3.652	.1314
Genotype 1 (vs. not 1)	1.602	0.657-3.907	.3004
Alcohol consumption <20 g/d (vs. ≥20 g/d)	0.431	0.203-0.913	.0280
Duration of the disease ≥20 y (vs. <20 y)	4.380	1.256-15.277	.0205
BMI ≥25.0 (vs. <25.0)	1.880	0.809-4.367	.1422
Comorbidities (vs. no comorbidities)	1.122	0.518-2.429	.7700
Diabetes (vs. no diabetes)	1.174	0.356-3.873	.7923
Hypertension (vs. no hypertension)	2.089	0.855-5.099	.1058
Fatty liver (vs. no fatty liver)	0.728	0.098-5.379	.7553
Cardiopulmonary disease (vs. no disease)	1.180	0.281-4.964	.8211

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder; BMI, body mass index (kg/m^2).

of the patients in our cohort died from cardiac disease (Table 9).

SMR in Relation to IFN Efficacy. SMR of the total cohort was 10.4 ($P < .01$) for liver-related death and 0.6 ($P < .05$) for liver-unrelated death (Table 10). SMR for all causes of death and liver-related death was lower in patients treated with IFN than in those untreated, mainly because of relatively low SMR in the sustained virologic responders. SMR in sustained virologic responders was reduced for liver-related death as well as liver-unrelated death. SMR for liver-unrelated death was less than 1.0 in both patients treated and untreated with IFN (Table 10).

SMR in Relation to Stage of Liver Fibrosis. SMR for all causes of death and liver-related death was increased gradually with progression from F0/1 to F4, whereas SMR for liver-unrelated death was less than 1.0 and remained unchanged from F0/1 to F3 except for stage F4 (Table 11). SMR for all causes of death was above 1.0 except for patients with stage F0/1, and SMR for liver-related death was extraordinarily higher in patients with F2 to F4 than in those with F0/1. IFN-treated patients showed lower SMR for all causes of death in each stage of F2 to F4 and for liver-related death in each stage of F0/1 to F4 than untreated patients (Table 11).

SMR in Relation to Age at Enrollment. SMR was calculated with the patients stratified according to age at enrollment. SMR for all causes of death and liver-related death was higher in the age group of 50 to 59 years than in the other age groups (Table 12). IFN treatment decreased SMR for all causes of death from 5.9 to 1.7 in the age group of 50 to 59 years, mainly because of the reduction of SMR for liver-related death from 45.5 to 9.6 (Table 12).

Discussion

This retrospective cohort study showed that IFN treatment provided a clinical advantage for patients with HCV-related chronic liver disease according to multivariate Cox regression analysis and SMR adjusted for an age- and sex-matched general population. Because this was a retrospective study and some of the baseline characteristics were different between IFN-treated and -untreated groups, our proportional hazards regression model used 15 variables potentially associated with survival including age, sex, fibrotic stage, and comorbidities to minimize the risk of uncontrolled confounding in the analysis.

Table 6. Risk for Liver-Related Death by Multivariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	*P* Value
Age			.3575
<49 y	1.0		
50-59 y	3.130	0.657-14.905	.1518
≥60 y	2.713	0.539-13.667	.2263
Fibrotic stage			.0088
F0/1	1.0		
F2	8.310	1.509-45.755	.0150
F3	12.866	2.323-71.253	.0034
F4	18.051	3.216-101.308	.0010
Interferon treatment			.0007
Untreated	1.0		
Treated	0.208	0.088-0.495	.0004
SVR	0.030	0.003-0.267	.0017
NSR	0.257	0.108-0.609	.0020
AST ≥80 IU/L (vs. <80)	10.170	2.289-45.176	.0023
ALT ≥80 IU/L (vs. <80)	0.291	0.062-1.365	.1174
Albumin ≥4.2 g/dL (vs. <4.2)	1.226	0.567-2.652	.6042
Platelet ≥13 × 10^9/L (vs. <13 × 10^9)	0.710	0.306-1.647	.4245
Alcohol consumption <20 g/d (vs. ≥20 g/d)	0.389	0.162-0.932	.0342
Duration of the disease ≥20 y (vs. <20 y)	1.554	0.388-6.219	.5334

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder.

Table 7. Risk for Liver-Unrelated Death by Univariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	P Value
Age			.0801
<49 y	1.0		
50-59 y	4.294	0.889-20.738	.0697
≥60 y	6.056	1.256-29.192	.0248
Male (vs. female)	4.942	1.119-21.827	.0350
Fibrotic stage			.0016
F0/1	1.0		
F2	1.601	0.293-8.748	.5868
F3	2.229	0.408-12.175	.3547
F4	9.384	2.794-31.52	.0003
Interferon treatment			.4433
Untreated	1.0		
Treated	1.972	0.447-8.693	.3694
SVR	1.304	0.218-7.818	.7711
NSR	2.294	0.507-10.374	.2807
AST ≥80 IU/L (vs. <80)	1.754	0.653-4.710	.2649
ALT ≥80 IU/L (vs. <80)	2.630	0.748-9.244	.1316
Albumin ≥4.2 g/dL (vs. <4.2)	0.283	0.102-0.781	.0149
Platelet ≥13 × 10^9/L (vs. <13 × 10^9)	0.350	0.131-0.934	.0360
HCV core protein ≥100 pg/mL (vs. <100)	0.998	0.359-2.772	.9965
Genotype 1 (vs. not 1)	2.384	0.538-10.570	.2528
Alcohol consumption <20 g/d (vs. ≥20 g/d)	0.596	0.192-1.852	.3706
Duration of the disease ≥20 y (vs. <20 y)	1.213	0.281-5.229	.7958
BMI ≥25.0 (vs. <25.0)	1.773	0.534-5.892	.3499
Comorbidities (vs. no comorbidities)	3.574	1.328-9.620	.0117
Diabetes (vs. no diabetes)	2.429	0.691-8.537	.1664
Hypertension (vs. no hypertension)	1.924	0.547-6.760	.3075
Fatty liver (vs. no fatty liver)	1.254	0.164-9.587	.8273
Cardiopulmonary disease (vs. no disease)	1.460	0.192-11.082	.7142

Abbreviations: SVR, sustained virologic responder; NSR, nonsustained responder; BMI, body mass index (kg/m^2).

There have been a number of studies on the mortality of patients with chronic hepatitis C, and several assessed the effect of IFN on their survival.[19-25] Niederau et al.[23] and Yoshida et al.,[25] using multivariate proportional regression analysis, reported that IFN treatment reduced the risk for overall death. Ikeda et al.[12] and Tanaka et al.[24] also reported that IFN treatment improved the survival rate, but they did not adjust their analysis for clinical factors. On the other hand, Fattovich et al.[21] and Gramenzi et al.,[22] both studies on cirrhotic patients, reported that IFN treatment had no benefit for survival. The present study showed that IFN treatment improved survival, especially in patients who responded to this therapy. As for cirrhotic patients, our results showed that IFN treatment reduced the risk of overall death to 0.348 (CI: 0.134-0.905) using multivariate Cox proportional hazards regression. All these studies reported proportions of liver-related to overall deaths of 57% to 94%, and, hence, a reduction in liver-related death is essential for improvement of survival in patients with chronic hepatitis C with or without cirrhosis. The proportions of deaths caused by HCC and liver failure to overall deaths, however, varied among these studies, from 13% to 33% vs. 31% to 63%,

Table 8. Risk for Liver-Unrelated Death by Multivariate Cox Regression Analysis

Variables	Risk Ratio	95% CI	P Value
Age			.2748
<49 y	1.0		
50-59 y	2.838	0.558-14.422	.2086
≥60 y	3.790	0.746-19.252	.1080
Male (vs. female)	6.063	1.353-27.177	.0185
Fibrotic stage			.0417
F0/1	1.0		
F2	0.871	0.151-5.020	.8769
F3	1.384	0.239-8.025	.7168
F4	5.020	1.289-19.546	.0200
Albumin ≥4.2 g/dL (vs. <4.2)	0.415	0.141-1.222	.1104
Platelet ≥13 × 10^9/L (vs. <13 × 10^9)	0.863	0.283-2.636	.7960
Comorbidities (vs. no comorbidities)	2.323	0.847-6.366	.1014

Table 9. SMR for Frequent Causes of Death in Patients With C-Viral Chronic Liver Disease

Cause of Death	No. of Deaths Observed	No. of Deaths Expected	SMR	P Value
All causes	48	30.95	1.6	<.01
Liver neoplasms	26	2.06	12.6	<.01
LC and CLD	6	1.02	5.9	<.01
Extrahepatic neoplasms	4	11.06	0.4	<.05
Cerebrovascular disease	5	3.41	1.5	NS
Cardiac disease	0	4.13	0	NS
Others	7	9.27	0.8	NS

NOTE. Liver neoplasms include HCC in 25 patients and cholangiocellular carcinoma in 1 patient. Extrahepatic neoplasms were seen in pancreas (n = 2), stomach (n = 1), and pyeloureter (n = 1). Other causes of death were the following: rupture of abdominal arterial aneurysm (n = 1), multiorgan failure (n = 1), traumatic brain damage (n = 1), suicide (n = 2), interstitial pneumonitis (n = 1), and fungal pneumonia (n = 1).

Abbreviations: LC, liver cirrhosis; CLD, chronic liver disease; NS, not significant.

Table 10. SMR for all Causes of Death and Liver-Related and -Unrelated Death in Relation to Interferon Efficacy in Patients With C-Viral Chronic Liver Disease

	All Causes of Death	Liver-Related Death	Liver-Unrelated Death
All cases	1.6 (48/30.95)*	10.4 (32/3.08)*	0.6 (16/27.87)†
IFN-untreated	2.0 (15/7.69)†	19.7 (13/0.66)*	0.3 (2/7.03)
IFN-treated	1.4 (33/23.26)	7.9 (19/2.42)*	0.7 (14/20.84)
SVR	0.6 (4/7.21)	1.3 (1/0.77)	0.5 (3/6.44)
NSR	1.8 (29/16.01)*	11.0 (18/1.64)*	0.8 (11/14.4)

NOTE. Liver disease–related causes of death include liver neoplasms, liver failure, and gastroesophageal variceal bleeding; liver disease–unrelated death includes all other causes. The numerator and denominator in parentheses show the number of observed deaths and expected deaths, respectively.

Abbreviations: SVR, sustained virologic responder, NSR, nonsustained responder.

*$P < .01$.

†$P < .05$.

Table 11. SMR for Liver-Related and -Unrelated Causes of Death in Relation to Stage of Liver Fibrosis in Patients With C-Viral Chronic Liver Disease

	All Causes of Death	Liver-Related Death	Liver-Unrelated Death
All patients, fibrotic stage			
F0/1	0.4 (6/14.03)*	1.4 (2/1.37)	0.3 (4/12.66)*
F2	1.5 (11/7.29)	12.3 (9/0.73)†	0.3 (2/6.56)
F3	2.4 (12/4.95)*	19.2 (10/0.52)†	0.5 (2/4.43)
F4	4.1 (19/4.68)†	24.4 (11/0.45)†	1.9 (8/4.23)
IFN-treated patients, fibrotic stage			
F0/1	0.5 (5/10.2)	1.0 (1/1.05)	0.4 (4/9.15)
F2	1.4 (8/5.56)	10.2 (6/0.59)†	0.4 (2/4.97)
F3	2.3 (10/4.4)*	19.0 (8/0.47)†	0.5 (2/3.93)
F4	3.2 (10/3.08)†	13.3 (4/0.30)†	2.2 (6/2.78)
Untreated patients, fibrotic stage			
F0/1	0.3 (1/3.82)	3.1 (1/0.32)	0 (0/3.50)
F2	1.7 (3/1.73)	21.43 (3/0.14)†	0 (0/1.59)
F3	3.6 (2/0.54)	40.0 (2/0.05)†	0 (0/0.49)
F4	5.7 (9/1.58)†	46.7 (7/0.15)†	1.4 (2/1.43)

NOTE. The numerator and denominator in parentheses show the number of observed deaths and expected deaths, respectively.

*$P < .05$.

†$P < .01$.

respectively, in European patients[21-23] and rather in contrast, from 45% to 54% vs. 11% to 14%, respectively, in Japanese patients[12,24,25] including ours. These differences can be ascribed to the diverse incidence rates of HCC in various countries, and Japan has a much higher rate than Europe, for as yet unexplained reasons.

SMR of patients with HCV-related chronic liver disease was 1.6, resulting in a slightly reduced life expectancy in the total cohort ($P < .01$). As for liver fibrosis, however, prognosis varied considerably among patients with different stages. Niederau et al.[23] and Yoshida et al.[25] reported that SMR of patients without cirrhosis was 0.9 and 0.8, respectively, and that noncirrhotic patients had a normal life expectancy. SMR of patients without cirrhosis (F0 to F3) in the current study was 1.1 (P = NS), and they seemed to have a normal life expectancy. But this result was influenced by the low SMR in patients with F0/1, and those with F2 and F3 had 1.5 and 2.4 times increased mortality for total death and 12.3 and 19.2 times increased mortality for liver-related death, respectively, when compared with rates expected for a matched general population. Thus, noncirrhotic patients with F2 and F3 need careful examinations and comprehensive treatment, including IFN therapy, as do cirrhotic patients, for improving their life expectancy. Indeed, IFN therapy reduced SMR from 5.7 to 3.2 (44% decrease), from 3.6 to 2.3 (36% decrease), and from 1.7 to 1.4 (18% decrease) in patients with F4, F3, and F2, respectively.

In our cohort, SMR for liver-related death was very high, whereas SMR for liver-unrelated death was less than 1.0. As patients with chronic hepatitis C generally have regularly scheduled medical examinations, the chance of early detection and management of liver-unrelated diseases might be elevated, or, on the other hand, patients with severe liver-unrelated disease who could not undergo liver biopsy might have been excluded from the study initially. Comorbidities such as diabetes mellitus, hypertension, fatty liver, cardiopulmonary diseases, and others were found in 26.6% of all patients, 25.4% in IFN-treated patients, and 30.8% in IFN-untreated patients in the current study ($P = .3125$). Liver-unrelated death was more frequently found in patients with comorbidity (7.4% vs. 2.1%, respectively, $P = .0166$). However, overall comorbidity was not associated with risk for liver-unrelated death by multivariate analysis (Table 8). SMR for liver-unrelated death was 1.9 in patients with cirrhosis (F4) when calculated with patients stratified according to fibrotic stage of the liver. Multivariate Cox regression analysis also showed fibrotic stage F4 as an independent risk factor for liver-unrelated death. Liver-unrelated diseases caused death in 8 (42%) of 19 cirrhotic patients, 6 (60%) of 10 treated with IFN, but in only 2 (22%) of 9 untreated patients. Thus, careful examination for liver-unrelated diseases as well as liver-related diseases will be necessary for patients with cirrhosis, especially for those treated with IFN.

Age at study entry might be one of the independent risk factors for overall death. Niederau et al.[23] reported that patients under 50 years at entry had an increased

Table 12. SMR for Liver-Related and -Unrelated Causes of Death in Relation to Age at Enrollment in Patients With C-Viral Chronic Liver Disease

	All Causes of Death	Liver-Related Death	Liver-Unrelated Death
All patients, age (y)			
-49	1.0 (4/3.82)	5.6 (2/0.36)	0.6 (2/3.46)
50-59	2.5 (27/10.71)*	15.6 (20/1.28)*	0.7 (7/9.43)
60-	1.0 (17/16.43)	6.9 (10/1.45)*	0.5 (7/14.98)†
IFN-treated patients, age (y)			
-49	1.3 (4/3.14)	6.7 (2/0.30)	0.7 (2/2.84)
50-59	1.7 (15/8.67)	9.6 (10/1.04)*	0.7 (5/7.63)
60-	1.2 (14/11.45)	6.5 (7/1.07)*	0.7 (7/10.38)
Untreated patients, age (y)			
-49	0 (0/0.68)	0 (0/0.06)	0 (0/0.62)
50-59	5.9 (12/2.04)†	45.5 (10/0.22)*	1.1 (2/1.82)
60-	0.6 (3/4.98)	7.9 (3/0.38)†	0 (0/4.60)

NOTE. The numerator and denominator in parentheses show the number of observed deaths and expected deaths, respectively.

*$P < .01$.

†$P < .05$.

SMR of 6.2, whereas those aged over 50 years had normal prognosis. In the present study, however, SMR was highest in the age group of 50 to 59 years, not of over 60 or under 50 years. The proportion of patients with advanced fibrosis (stage F3 or F4) was 13.8% in those under 50 years old but was 36.9% in those aged over 50 years, a difference contributing to the highest SMR in the 50 to 59 year age group. IFN treatment reduced SMR in this age group from 5.9 to 1.7 (71% reduction), and the most beneficial effect of IFN therapy might therefore be expected for patients in the age group of 50 to 59 years.

We were unable to determine the current survival status of 50 of the patients in the cohort because of their relocation. The analysis may have suffered from bias if some of them had died, in that they were assigned to the alive-status group at the date of their last confirmation of survival. However, the proportion of IFN-treated patients among the untraced ones was similar to that among the traced ones (40 of 50, 80% vs. 315 of 409, 77%, respectively, $P = .3927$), and the proportion of cirrhotic patients among the untraced patients was greater in the untreated group (2 of 10, 20% vs. 3 of 40, 7.5%, $P = .2581$). The mean age at entry among the untraced patients was older in the untreated group (51.8 ± 17.0 vs. 43.6 ± 13.0, respectively, $P = .1002$). Thus, the untreated group might have a higher risk of unrecorded overall death, especially liver-related deaths, and our current result might have underestimated the effect of IFN treatment on survival.

In conclusion, the current study showed increased SMR for overall death in patients with C-viral chronic liver disease but that IFN treatment decreased SMR mainly by a reduction in liver-related death. The decrease in SMR was most prominent in sustained virologic responders. In this study, a virologic sustained response was achieved in only 33% of 355 patients treated with IFN monotherapy. Recent progress in treatment with peginterferon combined with ribavirin showed elimination of serum HCV RNA in over 50% of treated patients,[32,33] with much higher rates of sustained virologic response. Based on our current results with standard IFN therapy, further improvement in the sustained virologic response rate from more effective antiviral drugs or combined therapy can be anticipated to provide normal life expectancy to patients with chronic hepatitis C.

References

1. Bruix J, Calvet X, Costa J, Ventura M, Bruguera M, Castillo R, Barrera JM, et al. Prevalence of antibodies to hepatitis C virus in Spanish patients with hepatocellular carcinoma and hepatic cirrhosis. Lancet 1989;2:1004-1006.
2. Tong MJ, El-Farra NS, Reikes AR, Co RL. Clinical outcome after transfusion-associated hepatitis C. N Engl J Med 1995;332:1463-1466.
3. Shiratori Y, Shiina S, Imamura M, Kato N, Kanai F, Okudaira T, Teratani T, et al. Characteristic difference of hepatocellular carcinoma between hepatitis B and C viral infection in Japan. HEPATOLOGY 1995;22:1027-1033.
4. Davis GL, Balart LA, Schiff ER, Lindsay K, Bodenheimer HC Jr, Perrillo RP, Carey W, et al. Treatment of chronic hepatitis C with recombinant interferon alfa. A multicenter randomized, controlled trial. Hepatitis Interventional Therapy Group. N Engl J Med 1989;321:1501-1506.
5. Shindo M, Di Bisceglie AM, Cristiano K, Feinstone SM, Hoofnagle JH. Decrease in serum hepatitis C viral RNA during alpha-interferon therapy for chronic hepatitis C. Ann Intern Med 1991;115:700-704.
6. Bonis PA, Ioannidis JP, Cappelleri JC, Kaplan MM, Lau J. Correlation of biochemical response to interferon alpha with histological improvement in hepatitis C: a meta-analysis of diagnostic test characteristics. HEPATOLOGY 1997;26:1035-1044.
7. Shiratori Y, Imazeki F, Moriyama M, Yano M, Arakawa Y, Yokosuka O, Kuroki T, et al. Histologic improvement of fibrosis in patients with hepatitis C who have sustained response to interferon therapy. Ann Intern Med 2000;132:517-524.
8. Reichard O, Foberg U, Fryden A, Mattsson L, Norkrans G, Sonnerborg A, Wejstal R, et al. High sustained response rate and clearance of viremia in chronic hepatitis C after treatment with interferon-alfa 2b for 60 weeks. HEPATOLOGY 1994;19:280-285.
9. Hagiwara H, Hayashi N, Mita E, Takehara T, Kasahara A, Fusamoto H, Kamada T. Quantitative analysis of hepatitis C virus RNA in serum during interferon alfa therapy. Gastroenterology 1993;104:877-883.
10. Shiratori Y, Kato N, Yokosuka O, Imazeki F, Hashimoto E, Hayashi N, Nakamura A, et al. Predictors of the efficacy of interferon therapy in chronic hepatitis C virus infection. Tokyo-Chiba Hepatitis Research Group. Gastroenterology 1997;113:558-566.
11. Imai Y, Kawata S, Tamura S, Yabuuchi I, Noda S, Inada M, Maeda Y, et al. Relation of interferon therapy and hepatocellular carcinoma in patients with chronic hepatitis C. Ann Intern Med 1998;129:94-99.
12. Ikeda K, Saitoh S, Arase Y, Chayama K, Suzuki Y, Kobayashi M, Tsubota A, et al. Effect of interferon therapy on hepatocellular carcinogenesis in patients with chronic hepatitis type C: a long-term observation study of 1643 patients using statistical bias correction with proportional hazard analysis. HEPATOLOGY 1999;29:1124-1130.
13. Yoshida H, Shiratori Y, Moriyama M, Arakawa Y, Ide T, Sata M, Inoue O, et al. Interferon therapy reduces the risk of hepatocellular carcinoma: national surveillance program of cirrhotic and noncirrhotic patients with chronic hepatitis C in Japan. Ann Intern Med 1999;131:174-181.
14. Nishiguchi S, Kuroki T, Nakatani S, Morimoto H, Takeda T, Nakajima S, Shiomi S, et al. Randomised trial of effects of interferon-alpha on incidence of hepatocellular carcinoma in chronic active hepatitis C with cirrhosis. Lancet 1995;346:1051-1055.
15. Mazzella G, Accogli E, Sottili S, Festi D, Orsini M, Salzetta A, Novelli V, et al. Alpha interferon treatment may prevent hepatocellular carcinoma in HCV-related liver cirrhosis. J Hepatol 1996;24:141-147.
16. Valla DC, Chevallier M, Marcellin P, Payen JL, Trepo C, Fonck M, Bourliere M, et al. Treatment of hepatitis C virus-related cirrhosis: a randomized, controlled trial of interferon alfa-2b versus no treatment. HEPATOLOGY 1999;29:1870-1875.
17. Fattovich G, Giustina G, Degos F, Diodati G, Tremolada F, Nevens F, Almasio P, et al. Effectiveness of interferon alfa on incidence of hepatocellular carcinoma and decompensation in cirrhosis type C. European concerted action on viral hepatitis (EUROHEP). J Hepatol 1997;27:201-205.
18. Camma C, Giunta M, Andreone P, Craxi A. Interferon and prevention of hepatocellular carcinoma in viral cirrhosis: an evidence-based approach. J Hepatol 2001;34:593-602.
19. Serfaty L, Aumaitre H, Chazouilleres O, Bonnand AM, Rosmorduc O, Poupon RE, Poupon R. Determinants of outcome of compensated hepatitis C virus-related cirrhosis. HEPATOLOGY 1998;27:1435-1440.
20. Benvegnu L, Chemello L, Noventa F, Fattovich G, Pontisso P, Alberti A. Retrospective analysis of the effect of interferon therapy on the clinical outcome of patients with viral cirrhosis. Cancer 1998;83:901-909.

21. Fattovich G, Giustina G, Degos F, Tremolada F, Diodati G, Almasio P, Nevens F, et al. Morbidity and mortality in compensated cirrhosis type C: a retrospective follow-up study of 384 patients. Gastroenterology 1997; 112:463-472.
22. Gramenzi A, Andreone P, Fiorino S, Camma C, Giunta M, Magalotti D, Cursaro C, et al. Impact of interferon therapy on the natural history of hepatitis C virus related cirrhosis. Gut 2001;48:843-848.
23. Niederau C, Lange S, Heintges T, Erhardt A, Buschkamp M, Hurter D, Nawrocki M, et al. Prognosis of chronic hepatitis C: results of a large, prospective cohort study. HEPATOLOGY 1998;28:1687-1695.
24. Tanaka H, Tsukuma H, Kasahara A, Hayashi N, Yoshihara H, Masuzawa M, Kanda T, et al. Effect of interferon therapy on the incidence of hepatocellular carcinoma and mortality of patients with chronic hepatitis C: a retrospective cohort study of 738 patients. Int J Cancer 2000;87:741-749.
25. Yoshida H, Arakawa Y, Sata M, Nishiguchi S, Yano M, Fujiyama S, Yamada G, et al. Interferon therapy prolonged life expectancy among chronic hepatitis C patients. Gastroenterology 2002;123:483-491.
26. Desmet VJ, Gerber M, Hoofnagle JH, Manns M, Scheuer PJ. Classification of chronic hepatitis: diagnosis, grading, and staging. HEPATOLOGY 1994;19:1513-1520.
27. Kato N, Yokosuka O, Hosoda K, Ito Y, Ohto M, Omata M. Quantification of hepatitis C virus by competitive reverse transcription-polymerase chain reaction: increase of the virus in advanced liver disease. HEPATOLOGY 1993;18:16-20.
28. Lau JY, Davis GL, Kniffen J, Qian KP, Urdea MS, Chan CS, MizokamiM, et al. Significance of serum hepatitis C virus RNA levels in chronic hepatitis C. Lancet 1993;341:1501-1504.
29. Tanaka E, Kiyosawa K, Matsumoto A, Kashiwakuma T, Hasegawa A, Mori H, Yanagihara O, et al. Serum levels of hepatitis C virus core protein in patients with chronic hepatitis C treated with interferon alfa. HEPATOLOGY 1996;23:1330-1333.
30. Tanaka T, Tsukiyama-Kohara K, Yamaguchi K, Yagi S, Tanaka S, Hasegawa A, Ohta Y, et al. Significance of specific antibody assay for genotyping of hepatitis C virus. HEPATOLOGY 1994;19:1347-1353.
31. Statistics and Information Department, Japan Ministry of Health and Welfare. Vital statistics in Japan (in Japanese). Tokyo: Health and Welfare Statistics Association, 1990, 1995, and 2000.
32. Glue P, Rouzier-Panis R, Raffanel C, Sabo R, Gupta SK, Salfi M, Jacobs S, et al. A dose-ranging study of pegylated interferon alfa-2b and ribavirin in chronic hepatitis C. HEPATOLOGY 2000;32:647-653.
33. Manns MP, McHutchison JG, Gordon SC, Rustgi VK, Shiffman M, Reindollar R, Goodman ZD, et al. Peginterferon alfa-2b plus ribavirin compared with interferon alfa-2b plus ribavirin for initial treatment of chronic hepatitis C: a randomised trial. Lancet 2001;358:958-965.