Keywords Hepatitis C virus · Normal alanine aminotransferase · Pegylated interferon plus ribavirin combination therapy · Propensity score method · Matched case–control study

Introduction

In patients with hepatitis C virus (HCV) infection, alanine aminotransferase (ALT) levels fluctuate and sometimes biochemical remission is maintained. Approximately 20% of patients with normal ALT levels (N-ALT) show ALT elevation and fibrosis progression within 3–5 years [1–5], and consequently, 70–80% of N-ALT patients have mild to moderate fibrosis on liver biopsy. N-ALT patients have been excluded from conventional interferon (IFN) therapy, because their sustained virological response (SVR) rates on conventional IFN monotherapy have been reported to be only 6–15% [6–9], and ALT levels were noted to increase during or after treatment in 47–62% of the patients. The incidence of ALT flares has raised concerns regarding the risk of conventional IFN therapy compared with a small benefit. However, a large randomized controlled trial has demonstrated that combination therapy with pegylated interferon (Peg-IFN) and ribavirin produced SVR rates in N-ALT patients with chronic hepatitis C (CHC) that were comparable to those of patients with elevated ALT levels (E-ALT) [10]. Thus, such treatment is now being considered for N-ALT patients with CHC [11].

Comparison of the characteristics of N-ALT and E-ALT patients has shown that the mean age of N-ALT patients was lower than that of E-ALT patients, and females and HCV genotype 2 patients were predominant among N-ALT patients [4, 7, 12–17]. In the American Association for the Study of Liver Disease guideline, the pretreatment predictors of achieving SVR with Peg-IFN plus ribavirin combination therapy for CHC patients are HCV genotype 2 or 3 infection, low viral load (<600 KIU/ml), female gender, and age less than 40 years [11]. Considering these characteristics, N-ALT patients with CHC can be said to have an advantageous background, and their response to antiviral therapy, including Peg-IFN plus ribavirin combination therapy, can be overestimated. Therefore, patient background, especially factors affecting the treatment efficacy of the combination therapy, needs to be matched between study groups in order to compare the treatment efficacies in N-ALT patients with CHC and E-ALT patients with CHC accurately. In this study, we evaluated, by a matched case–control study approach, whether the antiviral efficacy in N-ALT patients with CHC, reported to be equal to that in E-ALT patients with CHC, could be obtained without their advantageous background, and whether the factors contributing to SVR in N-ALT patients were the same as those in E-ALT patients. In addition, ALT flares after treatment in N-ALT patients without SVR were examined.

Patients and methods

Patient selection and study design

The subjects were 1320 consecutive CHC patients, 1015 with HCV genotype 1 (HCV-1) and 305 with HCV genotype 2 (HCV-2) who had undergone combination therapy with Peg-IFN alpha-2b (PEGINTRON; Schering-Plough, Kenilworth, NJ, USA) and ribavirin (REBETOL; Schering-Plough) at standard doses for 48 weeks (patients with HCV-1) or for 24 weeks (patients with HCV-2) at 30 medical institutions participating in the Osaka Liver Forum between December 2004 and December 2007. Peg-IFN alpha-2b and ribavirin dosages were based on body weight according to the manufacturer's instructions and were modified based on the manufacturer's instructions according to the severity of adverse hematologic effects. In the 1 month preceding treatment, none of the patients had received any IFN formulations or other types of drugs for liver supporting therapy. Before starting treatment, all patients had positive anti-HCV and a detectable level of HCV RNA according to a polymerase chain reaction (PCR)-based assay (COBAS Amplicor HCV Monitor Test v2.0; Roche Diagnostics, Branchburg, NJ, USA). None of the patients showed evidence of dual infection with hepatitis B virus or human immunodeficiency virus, or other forms of liver diseases such as alcoholic liver disorder, autoimmune hepatitis, or drug-induced liver injury.

In this study, a normal serum ALT level was defined as ALT $\leq$30 IU/l at the start of the combination therapy, as, in the guidelines for treatment of hepatitis C in N-ALT patients in Japan, ALT levels of $\leq$30 IU/l are regarded as an indicator of no or little inflammation in the liver, and patients whose ALT levels are $\leq$30 IU/l are recommended to be followed without antiviral therapy, especially if the platelet count is $\geq 15 \times 10^4/mm^3$.

Among the 1320 consecutive CHC patients, the antiviral effect in 193 N-ALT patients (116 with HCV-1, 77 with HCV-2) was compared with that in 193 E-ALT patients (116 with HCV-1, 77 with HCV-2) who were matched by a propensity score method based on age, sex, IFN treatment history, body mass index (BMI), and platelet counts. BMI was calculated as weight (kg)/[height (m)]^2.

HCV RNA was determined at week 4, week 12, end of treatment (EOT), and 24 weeks after EOT. HCV RNA was also determined at week 24 for HCV-1 patients. HCV RNA was monitored by the PCR Amplicor method with a detection limit of 50 IU/ml. (COBAS Amplicor HCV v2.0;

Roche Diagnostics). Complete early virological response (cEVR) and end-of-treatment response (ETR) were defined as undetectable HCV RNA at week 12 and EOT, respectively.

Written informed consent was obtained from each patient, and the study protocol was reviewed and approved according to the ethical guidelines of the 2004 Declaration of Helsinki by institutional review boards at the respective sites.

Propensity score

Propensity score methods are used to create balanced covariates and reduce selection bias in a matched case–control study. Propensity scores were calculated using a multivariate logistic regression model that had ALT levels as a dependent variable and other covariates as independent variables, and the model was utilized for matching between the N-ALT patients with CHC (the case group) and the E-ALT patients with CHC (the control group). Data analyses were conducted using SAS, version 9.2 (SAS Institute, Cary, NC, USA).

Statistical analysis

Continuous variables are reported as the mean with standard deviation (SD) or median levels, while categorical variables are shown as the count and proportion. Statistical significance was assessed by Student's t test (mean), the Mann–Whitney U test (median), and the χ^2 test for independent samples, and the paired t test for paired samples. For all tests, two-sided P values were calculated, and the results were considered statistically significant if $P < 0.05$. Variables that achieved statistical significance on univariate analysis were subjected to multivariate logistic regression analysis. Stepwise and multivariate logistic regression models were used to explore the independent factors that could be used to predict SVR. Statistical analysis was performed using the SPSS program for Windows, version 15.0 J (SPSS, Chicago, IL, USA).

Results

Baseline characteristics of all CHC patients according to HCV genotype and ALT levels before matching

The baseline characteristics of 1320 patients at the commencement of combination therapy with Peg-IFN and ribavirin are shown in Table 1, according to HCV genotype and ALT levels before matching. Of the 116 N-ALT patients with HCV-1 there were 36 males and 80 females (69%), with a mean age of 54 ± 11 years. Eighty-five (73%) were IFN-naïve. In terms of liver histology, 66 (73%) patients had

Table 1 Demographic characteristics of patients with normal ALT and patients with elevated ALT

	HCV genotype 1			HCV genotype 2		
	Normal ALT ($n = 116$)	Elevated ALT ($n = 899$)	P value	Normal ALT ($n = 77$)	Elevated ALT ($n = 228$)	P value
Sex: male/female	36/80	512/387	<0.001	32/45	121/107	0.081
Age (years)	54 ± 11	56 ± 10	0.136	51 ± 13	52 ± 13	0.423
Body mass index (kg/m^2)	22.9 ± 3.1	23.3 ± 3.2	0.131	23.0 ± 2.9	23.3 ± 3.2	0.424
Past IFN therapy: naïve/experienced (relapser/non-responder)[a]	85/31 (18/5)	547/352 (131/154)	0.011	58/19 (9/4)	175/53 (21/10)	0.876
Histology (METAVIR)[b]						
Activity: 0–1/2–3	66/25	296/338	<0.001	43/6	69/94	<0.001
Fibrosis: 0–1/2–4	67/24	330/304	<0.001	42/7	101/62	0.002
HCV RNA (KIU/ml)[c]	1800	1700	0.793	2200	1100	<0.001
White blood cell (/mm^3)	5220 ± 1507	5137 ± 1582	0.595	5538 ± 1687	5338 ± 1725	0.377
Neutrophil (/mm^3)	2770 ± 1074	2595 ± 1078	0.108	3017 ± 1180	2688 ± 1230	0.047
Hemoglobin (g/dl)	13.6 ± 1.5	14.2 ± 1.4	<0.001	13.8 ± 1.6	14.2 ± 1.4	0.071
Platelet (×10^4/mm^3)	19.9 ± 5.7	16.2 ± 5.3	<0.001	20.5 ± 4.5	17.8 ± 5.8	<0.001
ALT (IU/l)	24 ± 5	88 ± 62	<0.001	22 ± 5	97 ± 67	<0.001

ALT alanine aminotransferase, *HCV* hepatitis C virus, *IFN* interferon

[a] Status was unknown in 8 patients in the normal ALT group and 67 in the elevated ALT group with HCV genotype 1; and in 6 in the normal ALT group and 22 in the elevated ALT group with HCV genotype 2

[b] Data missing in 25 patients in the normal ALT group and in 265 in the elevated ALT group with HCV genotype 1; and in 28 in the normal ALT group and in 65 in the elevated ALT group with HCV genotype 2

[c] Values are expressed as medians

mild activity (activity, 0–1) and 67 (74%) had mild fibrosis (fibrosis, 0–1) by the METAVIR system. Mean white blood cell counts, hemoglobin levels, and platelet counts were 5220 ± 1570 /mm^3, 13.6 ± 1.5 g/dl, and 19.9 ± 5.7 × 10^4/mm^3. In 899 E-ALT patients compared to N-ALT patients, the proportions of female and IFN-naïve patients were significantly lower, at 43% ($P < 0.001$) and 61% ($P = 0.011$), respectively. Higher scores for activity ($P < 0.001$) and fibrosis ($P < 0.001$) were observed in E-ALT patients. E-ALT patients had higher hemoglobin levels and lower platelet counts than N-ALT patients, at 14.2 ± 1.5 g/dl ($P < 0.001$) and 16.2 ± 5.3 × 10^4/mm^3 ($P < 0.001$), respectively. Mean ALT levels were 24 ± 5 IU/l in N-ALT patients and 88 ± 62 IU/l in E-ALT patients ($P < 0.001$).

Of the 77 N-ALT patients with HCV-2, 32 were males and 45, females (58%). Their mean age was 51 ± 13 years and 58 (75%) were IFN-naïve. In terms of liver histology, 43 (88%) patients had mild activity (activity, 0–1) and 42 (86%) had mild fibrosis (fibrosis, 0–1). Compared to the 228 E-ALT patients, the N-ALT patients had higher HCV RNA levels (median 2200 vs. 1100 KIU/ml, $P < 0.001$). Higher scores for activity ($P < 0.001$) and fibrosis ($P = 0.002$) were observed in E-ALT patients. Neutrophils and platelet counts in N-ALT patients were higher than those in E-ALT patients, at 3017 ± 1180 versus 2688 ± 1230 /mm^3 ($P = 0.047$) and 20.5 ± 4.5 × 10^4 versus 17.8 ± 5.8 × 10^4/mm^3 ($P < 0.001$), respectively. Mean ALT levels were 22 ± 5 IU/l in N-ALT patients and 97 ± 67 IU/l in E-ALT patients ($P < 0.001$).

Prognostic factors for SVR in the N-ALT patients

For all N-ALT patients (HCV-1, 116; HCV-2, 77), univariate analysis for factors associated with achieving SVR was performed for the following variables: sex, age, BMI, history of past IFN therapy, histology, baseline HCV RNA level, HCV genotype, white blood cell count, neutrophil count, hemoglobin level, platelet count, and ALT level (Table 2). The results indicated that age, fibrosis, baseline HCV RNA level, and HCV genotype contributed to SVR. Next, multivariate logistic regression analysis was performed for all N-ALT patients ($n = 193$), using these factors except for fibrosis, as there were many missing samples. The multivariate analysis showed that younger age [by 10-year increase: odds ratio (OR) 0.552; 95% confidence interval (CI) 0.404–0.756; $P < 0.001$] and lower baseline HCV RNA level (by 100-KIU/ml increase: OR 0.976; 95% CI 0.954–0.998; $P = 0.037$), as well as HCV genotype (genotype 2 vs. genotype 1: OR 3.724; 95% CI 1.859–7.463; $P < 0.001$) were independently associated with SVR (Table 3).

Table 2 Factors associated with SVR in patients with normal ALT—univariate analysis

Factor	SVR ($n = 117$)	Non-SVR ($n = 76$)	P value
Sex: male/female	43/74	25/51	0.645
Age (years)	50 ± 13	57 ± 9	<0.001
Body mass index (kg/m^2)	22.8 ± 3.3	23.1 ± 2.6	0.511
Past IFN therapy: naïve/experienced	88/29	55/21	0.737
Histology (METAVIR)[a]			
Activity: 0–1/2–3	67/14	42/17	0.148
Fibrosis: 0–1/2–4	69/12	40/19	0.022
HCV genotype: 1/2	57/60	59/17	<0.001
HCV RNA (KIU/ml)[b]	1700	2100	0.040
White blood cell (/mm^3)	5461 ± 1426	5170 ± 1798	0.213
Neutrophil (/mm^3)	2968 ± 1167	2709 ± 1032	0.126
Hemoglobin (g/dl)	13.7 ± 1.4	13.7 ± 1.6	0.970
Platelet (×10^4/mm^3)	20.4 ± 4.8	19.8 ± 5.8	0.388
ALT (IU/l)	23 ± 5	24 ± 5	0.384

SVR sustained virological response, *ALT* alanine aminotransferase, *IFN* interferon, *HCV* hepatitis C virus

[a] Data missing in 36 patients in the SVR group and in 17 in the non-SVR group

[b] Values are expressed as medians

Table 3 Factors associated with SVR in patients with normal ALT—multivariate analysis

Factor	Category	Odds ratio	95% CI	P value
Age	By 10 years	0.552	0.404–0.756	<0.001
HCV genotype	1/2	3.724	1.859–7.463	<0.001
HCV RNA	By 100 KIU/ml	0.976	0.954–0.998	0.037

The number of patients used for this multivariate analysis was 193 (SVR, $n = 117$; non-SVR, $n = 76$)

SVR sustained virological response, *ALT* alanine aminotransferase, *CI* confidence interval, *HCV* hepatitis C virus

Comparison of patient characteristics between patients with normal ALT and those with elevated ALT matched by a propensity score method

The baseline characteristics of CHC patients matched by a propensity score method at the commencement of combination therapy with Peg-IFN and ribavirin were compared between N-ALT patients and E-ALT patients (see Table 4). There were 116 CHC patients with HCV-1 in each of the groups of N-ALT and E-ALT patients. The two groups were well matched by propensity score methods and there was no significant difference, except in ALT values (mean value, N-ALT, 24 ± 5 IU/l vs. E-ALT, 78 ± 53 IU/l, $P < 0.001$). Similarly, with CHC patients with HCV-2, there were no significant differences, except for ALT levels (mean value, N-ALT, 22 ± 5 IU/l vs. E-ALT, 80 ± 58 IU/l, $P < 0.001$), activity scores [0–1, N-ALT, 88% (43/49) vs. E-ALT, 49% (25/51), $P < 0.001$], and HCV RNA levels (median value, N-ALT, 2200 KIU/ml vs. E-ALT, 1000 KIU/ml, $P < 0.001$).

Treatment efficacy of combination therapy with Peg-IFN and ribavirin in CHC patients

Antiviral effects of the combination therapy with Peg-IFN and ribavirin were evaluated by rapid virological response (RVR), cEVR, ETR, SVR, and relapse rates, as shown in Table 5. Among patients with HCV-1 in the N-ALT and E-ALT patients, respectively, RVR rates were 6% (6/98) and 6% (6/102), cEVR rates were 53% (62/116) and 43% (50/116), and ETR rates were 72% (84/116) and 58% (67/116) ($P = 0.019$). SVR and relapse rates in N-ALT patients were 49% (57/116) and 32% (27/84). These rates in E-ALT patients were 40% (46/116) and 31% (21/67). In the patients with HCV-2, RVR, cEVR, ETR, SVR, and relapse rates were 68% (41/60), 90% (69/77), 96% (74/77), 78% (60/77), and 19% (14/74) for N-ALT patients, and 62% (36/58), 91% (70/77), 91% (70/77), 81% (62/77), and 11% (8/70) for E-ALT patients, respectively. Comparisons between N-ALT and E-ALT patients with HCV-1 or HCV-2 showed no significant differences in RVR, cEVR, ETR, SVR, and relapse rates, except in ETR rates in patients with HCV-1.

Changes in ALT levels during combination therapy and follow-up periods in N-ALT patients with SVR and those with non-SVR

Changes in ALT levels in N-ALT patients during the combination therapy and follow-up periods were evaluated according to the treatment response (Fig. 1). In patients with HCV-1, the mean baseline ALT level in the SVR

Table 4 Comparison of characteristics between patients with normal ALT and patients with elevated ALT matched by a propensity score method

	HCV genotype 1			HCV genotype 2		
	Normal ALT ($n = 116$)	Elevated ALT ($n = 116$)	P value	Normal ALT ($n = 77$)	Elevated ALT ($n = 77$)	P value
Sex: male/female	36/80	32/84	0.564	32/45	30/47	0.742
Age (years)	54 ± 11	55 ± 11	0.746	51 ± 13	50 ± 13	0.742
Body mass index (kg/m^2)	22.9 ± 3.1	22.6 ± 2.9	0.536	23.0 ± 2.9	22.8 ± 2.9	0.780
Past IFN therapy: naïve/experienced (relapser/non-responder)[a]	85/31 (18/5)	80/36 (13/18)	0.469	58/19 (9/4)	57/20 (7/4)	0.853
Histology (METAVIR)[b]						
Activity: 0–1/2–3	66/25	49/35	0.056	43/6	25/26	<0.001
Fibrosis: 0–1/2–4	67/24	59/25	0.736	42/7	36/15	0.068
HCV RNA (KIU/ml)[c]	1800	1700	0.896	2200	1000	<0.001
White blood cell (/mm^3)	5220 ± 1507	5329 ± 1626	0.569	5538 ± 1687	5530 ± 1780	0.977
Neutrophil (/mm^3)	2770 ± 1074	2702 ± 1094	0.641	3017 ± 1180	2755 ± 1189	0.189
Hemoglobin (g/dl)	13.6 ± 1.5	13.7 ± 1.4	0.542	13.8 ± 1.6	14.0 ± 1.4	0.592
Platelet (×10^4/mm^3)	19.9 ± 5.7	19.4 ± 7.1	0.562	20.5 ± 4.5	20.6 ± 5.5	0.911
ALT (IU/l)	24 ± 5	78 ± 53	<0.001	22 ± 5	80 ± 58	<0.001

ALT alanine aminotransferase, *IFN* interferon, *HCV* hepatitis C virus

[a] Data unknown in 8 patients in the normal ALT group and in 5 in the elevated ALT group with HCV genotype 1; and in 6 in the normal ALT group and 9 in the elevated ALT group with HCV genotype 2

[b] Data missing in 25 patients in the normal ALT group and in 32 in the elevated ALT group with HCV genotype 1; and in 28 in the normal ALT group and 26 in the elevated ALT group with HCV genotype 2

[c] Values are expressed as medians

Table 5 Antiviral effect for patients with normal ALT and those with elevated ALT according to HCV genotype

		Normal ALT	Elevated ALT	*P* value
HCV genotype 1		$n = 116$	$n = 116$	
	Undetectable HCV RNA rate			
	At week 4 (RVR)[a]	6% (6/98)	6% (6/102)	1.000
	At week 12 (cEVR)	53% (62/116)	43% (50/116)	0.287
	At week 48 (ETR)	72% (84/116)	58% (67/116)	0.019
	Post-24 weeks (SVR)	49% (57/116)	40% (46/116)	0.146
	Relapse rate	32% (27/84)	31% (21/67)	0.916
HCV genotype 2		$n = 77$	$n = 77$	
	Undetectable HCV RNA rate			
	At week 4 (RVR)[b]	68% (41/60)	62% (36/58)	0.563
	At week 12 (cEVR)	90% (69/77)	91% (70/77)	0.723
	At week 24 (ETR)	96% (74/77)	91% (70/77)	0.191
	Post-24 weeks (SVR)	78% (60/77)	81% (62/77)	0.691
	Relapse rate	19% (14/74)	11% (8/70)	0.212

ALT alanine aminotransferase, *HCV* hepatitis C virus, *RVR* rapid virological response, *cEVR* complete early virological response, *ETR* end-of-treatment response, *SVR* sustained virological response

[a] Data missing in 18 patients in the normal ALT group and in 14 in the elevated ALT group with HCV genotype 1

[b] Data missing in 17 patients in the normal ALT group and in 19 in the elevated ALT group with HCV genotype 2

group ($n = 57$) was similar to that in the non-SVR group ($n = 59$) (mean ± standard error of the mean (SEM): SVR group, 24.5 ± 0.6 IU/l; non-SVR group, 24.2 ± 0.7 IU/l; $P = 0.694$). Transitions of ALT levels were not significantly different between SVR and non-SVR groups during the therapy. However, in the SVR group, the ALT level fell to 15.1 ± 0.7 IU/l at 24 weeks after treatment completion ($P < 0.001$, compared to the baseline level), while in the non-SVR group, higher ALT levels were observed after treatment compared to the baseline level; the ALT level rose to the peak value of 36.2 ± 3.6 IU/l at post-12 weeks ($P = 0.001$), and slightly fell to 31.3 ± 2.6 IU/l at post-24 weeks ($P = 0.007$) (Fig. 1a). In comparison with the SVR group, the non-SVR group showed significant differences in mean ALT levels at post-4, -12, and -24 weeks ($P = 0.002$, <0.001, and <0.001, respectively). At post-48 weeks in the non-SVR group, the ALT level was 30.4 ± 2.9 IU/l, which was still higher than the baseline level ($P = 0.025$).

Similarly, in patients with HCV-2, baseline ALT levels in the SVR group ($n = 60$) and the non-SVR group ($n = 17$) were equivalent (mean ± SEM; SVR, 21.8 ± 0.7 IU/l; non-SVR, 22.5 ± 1.1 IU/l; $P = 0.622$), and there was no significant difference in transitions of the ALT levels during therapy. However, after treatment, in the non-SVR group, ALT levels tended to rise in comparison with those at baseline; they rose to 74.9 ± 26.9 IU/l at post-12 weeks ($P = 0.068$) and fell to 35.7 ± 10.2 IU/l at post-24 weeks ($P = 0.196$). On the other hand, in the SVR group, ALT levels fell significantly, to 16.4 ± 1.3 IU/l at post-12 weeks ($P < 0.001$) and 15.2 ± 1.2 IU/l at post-24 weeks ($P < 0.001$) (Fig. 1b). Comparison of ALT levels between the SVR and non-SVR groups after treatment showed that mean ALT levels in the non-SVR group tended to be high at post-4, -12 and, -24 weeks ($P = 0.045$, 0.051, and 0.066, respectively). At post-48 weeks in the non-SVR group, the ALT level was 32.4 ± 8.9 IU/l, which tended to be high compared with the baseline ALT level, although no significant difference was found ($P = 0.248$).

Next, the ALT levels in N-ALT patients were examined according to the treatment response at 24 weeks after completion of the combination therapy. In HCV-1 patients with SVR, ALT levels remained below the upper limit of normal (ULN) for this study (<30 IU/l) in 55 (98%) patients, and ALT elevation <2 × ULN occurred in only one (2%) patient (ALT 32 IU/l). On the other hand, in patients with non-SVR, ALT levels remained stable in 34 (60%) patients but increased to <2 × ULN in 20 (35%) patients, and to ≥2 × ULN in 3 (5%) patients (ALT 62, 79, and 135 IU/l). Similarly, in HCV-2 patients with SVR, ALT levels remained stable in 56 (95%) patients, and ALT elevation rarely occurred [<2 × ULN, 2 (3%) patients; ≥2 × ULN, one (2%) patient (ALT 68 IU/l)]. In contrast, in patients with non-SVR, ALT levels remained normal in 10 (67%) patients but increased to <2 × ULN in 4 (27%) patients and to ≥2 × ULN in one (6%) patient (ALT 174 IU/l).

Discussion

N-ALT patients with CHC are known to show demographic and virological features associated with higher

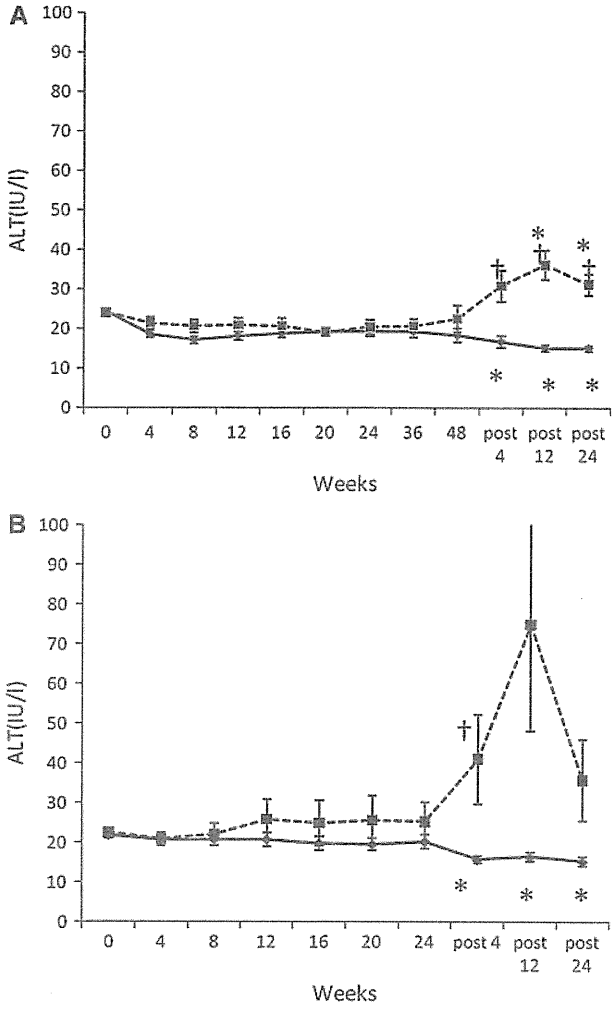

Fig. 1 Changes in serum alanine aminotransferase (*ALT*) levels (±standard error of the mean) according to response in patients with normal ALT levels with chronic hepatitis C treated with pegylated interferon and ribavirin. *Solid lines* show ALT levels in patients with a sustained virological response (SVR), and *dashed lines* show these levels in patients with a non-SVR. *Single-asterisks* denote a statistically significant difference ($P < 0.05$) in mean ALT levels between baseline and each time point of the follow-up period. *Daggers* denote a statistically significant difference between SVR and non-SVR groups. a Patients infected with hepatitis C virus genotype 1 (HCV-1). The number of patients was 57 in the SVR group and 59 in the non-SVR group. b HCV-2 patients. The number of patients was 60 in the SVR group and 17 in the non-SVR group

response rates to Peg-IFN and ribavirin combination therapy [4, 7, 12–17]. In the present study, N-ALT patients were younger and had higher platelet counts than E-ALT patients, thus giving N-ALT patients an advantage in antiviral efficacy in comparison with E-ALT patients in our cohort. However, the preponderance of females was greater in N-ALT patients with HCV-1 in this study, giving N-ALT patients a disadvantage. Accordingly, a direct comparison was made between these two patient groups after matching E-ALT patients with N-ALT patients using propensity score methods to reduce the bias due to differences in patient backgrounds. As a result, the efficacy of the combination therapy in N-ALT patients was revealed to be still equivalent to that in E-ALT patients, irrespective of their advantageous background. Moreover, in N-ALT patients with HCV-1, not only the ETR rate, but also the SVR rate tended to be higher than these rates in E-ALT patients (49% in N-ALT patients vs. 40% in E-ALT patients). Accordingly, N-ALT patients with HCV-1 can achieve a better treatment response in comparison with E-ALT patients, but further study is needed to clarify this.

In the present study, multivariate logistic regression analysis showed that achieving SVR was strongly influenced by HCV genotype and baseline HCV RNA level in N-ALT patients, which was consistent with findings of multicenter studies with E-ALT patients [18–21]. Therefore, decisions for treatment and the treatment regimen for N-ALT patients can mirror those recommended for E-ALT patients. The results of our multivariate analysis also revealed that patient age influenced the achievement of SVR in N-ALT patients. This offers support for the decision to offer antiviral treatment to younger N-ALT patients.

Among patients in our study who achieved SVR with the combination therapy, ALT levels after treatment decreased significantly, as shown in Fig. 1. However, approximately 40% of the non-SVR patients had increased ALT levels of up to $<2 \times$ ULN, and about 5% of patients had increased ALT levels of $\geq 2 \times$ ULN at 24 weeks after completion of the combination therapy, regardless of HCV genotype. When N-ALT patients are commencing the combination therapy, these patients should be told about the possibility of ALT exacerbation [6–9], although it is difficult to know whether this is drug-induced or due to the natural course. It is also difficult to state which patient characteristics make ALT elevation more likely to occur after the treatment.

Taking the findings obtained in the present study together, in N-ALT patients with HCV genotype 2, earlier treatment with Peg-IFN plus ribavirin combination therapy is desirable, as better efficacy was found for younger patients, with an SVR rate of approximately 80% being attained with this combination therapy, and few direct-acting antiviral agents (DAAs) have been developed for genotype 2. On the other hand, N-ALT patients with HCV genotype 1 should consider awaiting the DAAs, because SVR cannot be attained in about half of these patients, and the ALT level rises after treatment in about 40% of patients with non-SVR.

From the aspect of long-term prognosis, we need to verify, by prospective study, that viral eradication is really required for N-ALT patients because the incidence of hepatocellular carcinoma and liver-related mortality in

N-ALT patients has not been clarified. Deuffic-Burban et al. [22] calculated the impact of Peg-IFN plus ribavirin on morbidity and mortality in N-ALT patients using the Markov model and concluded that antiviral therapy in N-ALT patients would decrease morbidity and mortality rates. However, the treatment of N-ALT patients with CHC still remains an area of investigation, particularly with respect to the benefit-to-risk ratio of treatment. To help determine the indications for antiviral therapy in N-ALT patients, the liver histology should be evaluated before treatment. The presence of significant hepatic fibrosis (≥F2 by the METAVIR classification [23]) reflects continuous hepatic inflammation over a period of time and suggests a future risk of liver-related disease progression. Antiviral therapy may be appropriate for these patients. On the other hand, periodic follow up without antiviral therapy is recommended for patients in stages F0-1, because most of such patients show a low risk for progression to cirrhosis and the development of hepatocellular carcinoma [24].

This study had some limitations. First, the factors of viral mutation and host genetic mutation, which have been reported recently to affect the efficacy of Peg-IFN plus ribavirin combination therapy, could not be measured, and evaluation of the serum HCV RNA levels by a real-time PCR method, which is more sensitive to the measurement of serum HCV RNA levels, could not be done in the patients enrolled in this study, because we had few stored patient serum samples. Detailed examinations using the real-time PCR method in patients who are matched based on the factors of viral mutation and host genetic mutation as well as background factors will be needed for further study. Second, we excluded the factor of fibrosis from the multivariate analysis for factors associated with SVR in N-ALT patients, because data for fibrosis were lacking in 53 of the 193 patients in this study. Accordingly, the present study could not demonstrate whether fibrosis was associated with SVR in N-ALT patients. Finally, in this study, we investigated the antiviral efficacy of Peg-IFN plus ribavirin combination therapy for patients with N-ALT at the start of the therapy, not for patients with 'persistently' normal ALT. Accordingly, this study does not show the efficacy of this treatment in patients with persistently normal ALT. However, we believe that the results obtained in this study can be useful for pre-treatment prediction in outpatients who may not be followed by the reason of having normal ALT levels.

We have shown, in this matched case–control study using a propensity score method, that the therapeutic effect of combination therapy with Peg-IFN alpha-2b and ribavirin in N-ALT patients with CHC is comparable to that for E-ALT patients, irrespective of their advantageous background. Further work is needed to verify that HCV eradication can improve the prognosis of N-ALT patients.

Acknowledgments Other institutions and participants in the Osaka Liver Forum are: Higashi Osaka General Hospital, S. Iio; Sumitomo Hospital, A. Yamada; Osaka General Medical Center, A. Inoue; Toyonaka Municipal Hospital, M. Inada; National Hospital Organization Osaka Minami Medical Center, T. Hijioka; Yao Municipal Hospital, H. Fukui; Kinki Central Hospital of Mutual Aid Association of Public School Teachers, E. Hayashi; Osaka Koseinenkin Hospital, T. Ito; Itami City Hospital, T. Kashihara; Suita Municipal Hospital, T. Nagase; Ashiya Municipal Hospital, K. Kiriyama; Saiseikai Senri Hospital, K. Suzuki; NTT West Osaka Hospital, A. Kaneko; National Hospital Organization Minami Wakayama Medical Center, M. Kato; Otemae Hospital, Y. Doi; Kano General Hospital, S. Kubota; Nishinomiya Municipal Central Hospital, H. Ogawa; and Osaka Kaisei Hospital, N. Imaizumi. This work was supported by a Grant-in-Aid for Research on Hepatitis and BSE from the Ministry of Health, Labour and Welfare of Japan, and Scientific Research from the Ministry of Education, Science, and Culture of Japan. All authors have no financial relationship relevant to this study to disclose.

Conflict of interest The authors declare that they have no conflict of interest.

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J Gastroenterol (2011) 46:1031–1037
DOI 10.1007/s00535-011-0409-7

ORIGINAL ARTICLE—LIVER, PANCREAS, AND BILIARY TRACT

Efficacy of re-treatment with pegylated interferon plus ribavirin combination therapy for patients with chronic hepatitis C in Japan

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Received: 27 December 2010 / Accepted: 31 March 2011 / Published online: 3 May 2011

Abstract
Background It is still not known which patients with chronic hepatitis C who failed to respond to previous pegylated interferon (Peg-IFN) plus ribavirin therapy can benefit from re-treatment.
Methods Seventy-four patients (HCV genotype 1, $n = 56$, genotype 2, $n = 18$) were re-treated with Peg-IFN plus ribavirin.
Results On re-treatment, the sustained virologic response (SVR) rate was 41% for genotype 1 and 56% for genotype 2. With genotype 1, the factors associated with an SVR were previous treatment response and the serum hepatitis C virus (HCV) RNA level at the start of re-treatment. Patients with a ≥2-log decrease in HCV RNA at week 12 (partial early virologic response, p-EVR) in previous treatment had significantly higher SVR rates than those without these

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decreases ($p < 0.001$); no patient without a p-EVR in the previous treatment attained an SVR with re-treatment (0/16). All patients with <5 $\log_{10}$ IU/ml of HCV RNA at the start of re-treatment attained an SVR (6/6), while only 33% (15/45) of those patients with ≥5 $\log_{10}$ IU/ml of HCV RNA attained an SVR ($p < 0.01$). Among the patients with relapse in the previous treatment, those who attained an SVR on re-treatment required a longer duration of re-treatment than the duration of the previous treatment (re-treatment, 63.8 ± 13.0 weeks vs. previous treatment, 53.9 ± 13.5 weeks, $p = 0.01$).

Conclusions Re-treatment of genotype 1 patients should be limited to patients with a p-EVR in the previous treatment and a low HCV RNA level at the start of re-treatment. In re-treatment with Peg-IFN plus ribavirin, longer treatment duration can contribute to increasing the anti-viral effect.

Keywords Chronic hepatitis C · Pegylated interferon and ribavirin combination therapy · Re-treatment

Introduction

Pegylated interferon (Peg-IFN) plus ribavirin combination therapy can improve anti-viral efficacy and is currently recommended as first-line therapy for chronic hepatitis C. However, hepatitis C virus (HCV) still persists in approximately half of the genotype 1 patients treated with Peg-IFN plus ribavirin [1–4], and the number of patients who fail to achieve a sustained virologic response (SVR) consequently increases over time.

Recently, the addition of a protease inhibitor to Peg-IFN plus ribavirin combination therapy has been reported to improve the anti-viral effect, but this triple therapy increases side effects, especially severe anemia [5–7]. In Japan, HCV carriers are 10–20 years older than those in the United States and European countries, and patients who are ineligible for triple therapy exist in large numbers due to their potential tendency of having anemia. On the other hand, re-treatment with Peg-IFN plus ribavirin is a possible choice, until triple therapy becomes commercially available, for patients who have failed to show an SVR to previous anti-viral therapy, and for patients who are ineligible for triple therapy. As for re-treatment with Peg-IFN plus ribavirin, there have been only a few studies of patients who failed to show an SVR to previous Peg-IFN plus ribavirin [8–11]. Although re-treatment with Peg-IFN plus ribavirin for patients who failed to respond to previous Peg-IFN plus ribavirin is not recommended in the practice guidelines of the American Association for the Study of the Liver (AASLD) [1], there are some patients who respond to re-treatment. However, it remains obscure in which patients eradication of HCV can be successfully attained by re-treatment with Peg-IFN plus ribavirin.

In the present study, we tried to determine which patients could benefit from re-treatment and to identify the factors associated with an SVR in re-treatment.

Patients and methods

Patients

The present study was a retrospective, multicenter trial conducted by Osaka University Hospital and other institutions participating in the Osaka Liver Forum. This study was conducted with 74 chronic hepatitis C patients (genotype 1, $n = 56$, genotype 2, $n = 18$) who had previously completed Peg-IFN α-2b plus ribavirin combination therapy but had failed to attain an SVR. Patients were excluded from this study if they had decompensated cirrhosis or other forms of liver disease (alcoholic liver disease, autoimmune hepatitis), or coinfection with hepatitis B or anti-human immunodeficiency virus. This study was conducted according to the ethical guidelines of the Declaration of Helsinki amended in 2008, and informed consent was obtained from each patient.

Treatment

For the previous treatment, Peg-IFN α-2b (Pegintron; Schering-Plough, Kenilworth, NJ, USA) plus ribavirin (Rebetol; Schering-Plough) was started between December 2004 and January 2008. For re-treatment with Peg-IFN plus ribavirin, Peg-IFN α-2a (Pegasys; Roche, Basel, Switzerland) plus ribavirin (Copegus; Roche) or Peg-IFN α-2b plus ribavirin was started between February 2006 and January 2009. In principle, as a starting dose, Peg-IFN was given once weekly at a dose of 180 μg of Peg-IFN α-2a and 1.5 μg/kg of Peg-IFN α-2b, and ribavirin was given at a total dose of 600–1000 mg/day based on body weight (for genotype 1, body weight <60 kg, 600 mg; 60–80 kg, 800 mg; >80 kg, 1000 mg; for genotype 2, body weight <60 kg, 600 mg; >60 kg, 800 mg), according to a standard treatment protocol for Japanese patients.

Dose reduction and discontinuance

Dose modification followed, as a rule, the manufacturer's drug information on the intensity of the hematologic adverse effects. The Peg-IFN α-2a and α-2b doses were reduced to 50% of the assigned dose when the neutrophil count fell below 750/mm^3 or the platelet (Plt) count fell below 8×10^4/mm^3, and the agent was discontinued when the neutrophil count fell below 500/mm^3 or the Plt count fell below 5×10^4/mm^3. Ribavirin was also reduced from 1000 to 600, 800 to 600, or 600 to 400 mg when the

hemoglobin (Hb) was below 10 g/dl, and was discontinued when the Hb was below 8.5 g/dl. Both Peg-IFN and ribavirin had to be discontinued if there was a need to discontinue one of the drugs. No iron supplement or hematopoietic growth factors, such as epoietin alpha or granulocyte–macrophage colony stimulating factor (G-CSF), were administered.

Virologic assessment and definition of virologic response

The serum HCV RNA level was quantified using the COBAS AMPLICOR HCV MONITOR test, version 2.0 (detection range 6–5000 KIU/ml; Roche Diagnostics, Branchburg, NJ, USA) and qualitatively analyzed using the COBAS AMPLICOR HCV test, version 2.0 (lower limit of detection 50 IU/ml). A rapid virologic response (RVR) was defined as undetectable serum HCV RNA level at week 4, a partial early virologic response (p-EVR) was defined as more than a 2-log decrease in HCV RNA level at week 12 compared with the baseline, a complete EVR (c-EVR) was defined as undetectable serum HCV RNA at week 12, a late virologic response (LVR) was defined as detectable serum HCV RNA at week 12 and undetectable at week 24, and an SVR was defined as undetectable serum HCV RNA at 24 weeks after the end of the treatment. Relapse was defined as undetectable serum HCV RNA at the end of the treatment but a detectable amount after the end of the treatment. For both the previous treatment and this re-treatment, patients without a p-EVR or without clearance of HCV RNA at week 24 were considered to be showing non-response (NR) and had to stop treatment. A patient who attained HCV RNA negativity during the re-treatment continued to be treated for 48 or 72 weeks according to response-guided therapy and the decision of the investigator at the participating clinical center.

Statistical analysis

Baseline data of the patients are expressed as mean ± SD or median values. In order to analyze the differences between baseline data or the factors associated with SVR, univariate analysis using the Mann–Whitney *U*-test or the χ^2 test was performed. A two-tailed p value of <0.05 was considered significant. The analysis was conducted with SPSS version 15.0J (SPSS, Chicago, IL, USA).

Results

The baseline characteristics of the patients are summarized in Table 1. Of the 56 genotype 1 patients, 32 were relapsers and 24 showed NR to previous treatment. Among the relapsers, 15 had shown a c-EVR (58%, 15/26) and 29 a p-EVR (100%, 29/29) in the previous treatment. Of the 18 genotype 2 patients, 17 were relapsers and one had shown NR to the previous treatment. Among the relapsers, 5 had shown an RVR (42%, 5/12) in the previous treatment. In the previous treatment, all patients had received Peg-IFN α-2b plus RBV combination therapy. There were no significant differences among the baseline characteristics between the previous treatment and the re-treatment in

Table 1 Baseline characteristics of patients and treatment factors in previous treatment and re-treatment

	Genotype 1						Genotype 2	
	All patients		Previous treatment relapsers		Previous treatment non-responders		All patients	
Number of patients	56		32		24		18	
Sex: male/female	32/24		19/13		13/11		11/7	
	Previous treatment	Re-treatment	Previous treatment	Re-treatment	Previous treatment	Re-treatment	Previous treatment	Re-treatment
Age (years)	57.6 ± 9.2	59.5 ± 9.4	57.8 ± 9.0	59.8 ± 9.4	57.3 ± 9.6	59.0 ± 9.5	57.4 ± 9.0	58.4 ± 1.7
White blood cells (/mm^3)	4909 ± 1404	4670 ± 1566	5117 ± 1276	4756 ± 979	4633 ± 1543	4545 ± 2178	5111 ± 1697	4412 ± 1744
Red blood cells (×10^4/mm^3)	435 ± 40	426 ± 52	444 ± 34	437 ± 36	4243 ± 46	412 ± 67	448 ± 36	447 ± 38
Hemoglobin (g/dl)	13.9 ± 1.2	13.5 ± 1.7	14.1 ± 1.1	13.8 ± 1.3	13.7 ± 1.3	13.1 ± 2.1	14.4 ± 1.2	14.2 ± 1.3
Platelets (×10^4/mm^3)	16.5 ± 6.1	17.5 ± 6.9	18.4 ± 6.6	19.1 ± 6.5	14.1 ± 4.4	15.2 ± 6.9	17.5 ± 6.3	16.2 ± 4.9
AST (IU/l)	58 ± 30	60 ± 45	55 ± 31	56 ± 44	61 ± 28	64 ± 47	52 ± 34	34 ± 13
ALT (IU/l)	74 ± 55	77 ± 74	73 ± 65	79 ± 80	74 ± 40	75 ± 66	65 ± 52	34 ± 18
Serum HCV RNA (KIU/ml)	1600	1100	1600	1100	1600	990	1300	690
Peg-IFN type: α2a/α2b	0/56	24/32	0/32	14/18	0/24	10/14	0/18	4/14

AST aspartate aminotransferase, *ALT* alanine aminotransferase, *HCV* hepatitis C virus, *Peg-IFN* pegylated interferon

Table 2 Factors associated with a sustained virologic response (SVR) in re-treatment with Peg-IFN plus ribavirin

Factor	SVR	Non-SVR	*p* value
Number of patients	23	33	
Age (years)	59.5 ± 7.6	59.5 ± 10.5	0.55
Sex: male/female	16/7	16/17	0.17
White blood cells (/mm^3)	4778 ± 1022	4589 ± 1884	0.29
Neutrophils (/mm^3)	2446 ± 849	2291 ± 1486	0.21
Hemoglobin (g/dl)	13.6 ± 1.3	13.4 ± 1.9	0.73
Platelets (×10^4/mm^3)	18.2 ± 6.3	16.9 ± 7.3	0.28
AST (IU/l)	52 ± 33	65 ± 52	0.46
ALT (IU/l)	75 ± 61	79 ± 82	0.72
Serum HCV RNA: <5log/5log≤	6/15	0/31	<0.01
Peg-IFN type: α2a/α2b	7/16	17/16	0.27
Peg-IFN dose (μg/kg/week)			
α2a	2.64 ± 0.61	2.73 ± 0.72	0.90
α2b	1.18 ± 0.43	1.19 ± 0.34	0.90
Ribavirin dose (mg/kg/day)	8.6 ± 2.9	9.4 ± 2.7	0.28
1st treatment virologic response			
p-EVR; +/−	22/0	14/16	<0.001
Relapse/NR	20/3	12/21	<0.001

p-EVR partial early virologic response, *NR* non-response

terms of peripheral blood cell counts, or the levels of aminotransaminases and serum HCV RNA at the start of treatment.

In genotype 1 patients, the HCV RNA negative rate on re-treatment was 54% (29/54) at week 12 and 71% (40/56) at week 24, and the SVR rate was 41% (23/56). The factors associated with SVR were assessed by univariate analysis for the following variables; age, gender, peripheral blood cell counts, aminotransferases, previous treatment response, serum HCV RNA level, the type of Peg-IFN in re-treatment, and drug adherence (Table 2). As a result, the factors of previous treatment response and serum HCV RNA level at the start of re-treatment were selected as being significant. In examining the efficacy of the re-treatment according to the previous treatment response, the relapsers in the previous treatment had a significantly higher HCV RNA negative rate at weeks 12 and 24 and a significantly higher SVR rate than those with NR in the previous treatment (Fig. 1a). Patients with a p-EVR in the previous treatment showed similar results, while no patient without p-EVR in the previous treatment attained an SVR on re-treatment (0/16) (Fig. 1b). Even among the patients without HCV RNA negativity in the previous treatment, if p-EVR had been attained in the previous treatment, 43% (3/7) of these patients attained an SVR on re-treatment. As for the serum HCV RNA level at the start of re-treatment, all patients with less than 5 $\log_{10}$ IU/ml of HCV RNA attained an SVR (6/6), and 33% (15/45) of those patients with more than 5 $\log_{10}$ IU/ml of HCV RNA attained an SVR ($p < 0.01$).

In examining the efficacy of re-treatment according to treatment duration, among the patients with c-EVR and without RVR on re-treatment, those who were re-treated for 72 weeks tended to attain higher SVR rates than those who were re-treated for 48 weeks (72 weeks, 75%, 9/12, vs. 48 weeks, 25%, 2/8, $p = 0.06$). On the other hand, 43% (3/7) of the patients with an LVR on re-treatment attained an SVR on re-treatment. Among the patients with relapse

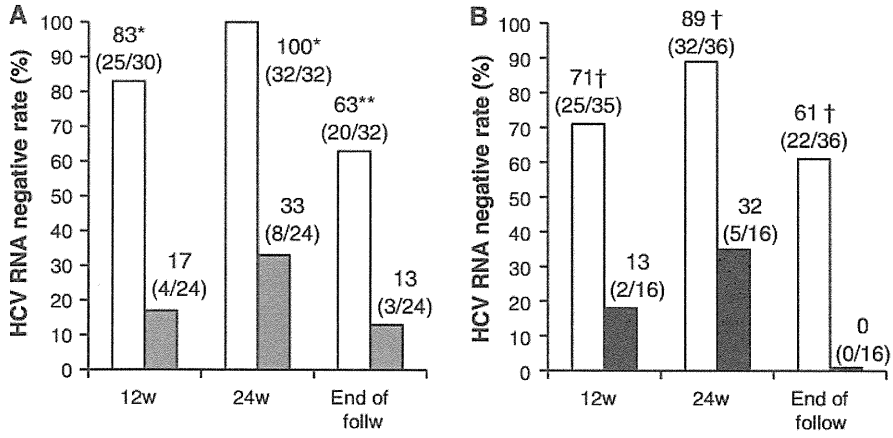

Fig. 1 Virologic response on re-treatment according to previous treatment response. **a** Hepatitis C virus (HCV) RNA negative rate on re-treatment according to relapse or non-response in previous treatment. **b** HCV RNA negative rate on re-treatment according to partial early virologic response (p-EVR) or non-p-EVR in previous treatment. *White bars* patients with relapse in previous treatment. *Dark gray bars* patients with non-response in previous treatment. *Light gray bars* patients with p-EVR in previous treatment. *Black bars* patients with non-p-EVR in previous treatment. *$p < 0.0001$; **$p < 0.01$; compared to non-response. †$p < 0.001$; compared to patients without p-EVR

in the previous treatment, those who attained an SVR on re-treatment required a longer duration of re-treatment than the duration of the previous treatment (re-treatment, 63.8 ± 13.0 weeks vs. previous treatment, 53.9 ± 13.5 weeks, $p = 0.01$), while those without an SVR on re-treatment could be treated for almost the same period as that in the previous treatment (re-treatment, 58.8 ± 12.8 weeks vs. previous treatment, 54.2 ± 11.3 weeks, $p = 0.38$).

Comparison of the timing to the first undetectable HCV RNA level in the previous treatment and re-treatment could be carried out in 50 patients; most patients attained HCV RNA negativity on re-treatment earlier or with the same timing as in the previous treatment, and only one patient showed a later timing for re-treatment. The SVR rate on re-treatment was low, at 13% (3/24) among the patients with detectable HCV RNA at week 24 in the previous treatment. Among the 10 patients with HCV RNA negativity on re-treatment with the same timing as that in the previous treatment, an SVR was attained only by the patients who were re-treated for 72 weeks. Among the 23 patients with earlier HCV RNA negativity on re-treatment, an SVR of 61% was attained (14/23). The patients with an RVR on re-treatment attained a high SVR rate (88%, 7/8) regardless of the virologic response in the previous treatment (Fig. 2).

In genotype 2 patients, the HCV RNA negative rate on re-treatment was 56% (10/18) at week 4, 83% (15/18) at week 12, and 89% (16/18) at week 24, and the SVR rate was 56% (10/18). The two patients without a c-EVR in the previous treatment did not attain an SVR on re-treatment. Among the patients with an RVR on re-treatment, the SVR rates were 60% (3/5) in those with 24-week treatment and 100% (5/5) in those with 48-week treatment.

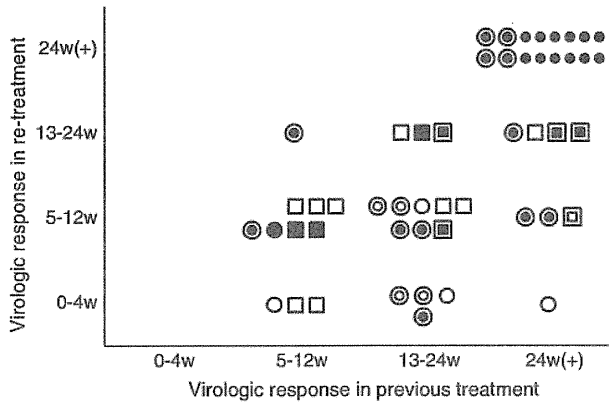

Fig. 2 Virologic response on re-treatment according to the timing of HCV RNA negativity in previous treatment and re-treatment. *Open double circles/open circles* sustained virologic response (SVR) with 48 weeks of re-treatment (*open double circles*, pegylated interferon [Peg-IFN] α-2a plus ribavirin; *open circles* Peg-IFN α-2b plus ribavirin). *Open double squares/open squares*, SVR with 72 weeks of re-treatment (*open double squares* Peg-IFN α-2a plus ribavirin; *open squares*, Peg-IFN α-2b plus ribavirin). *Closed double circles/closed circles*, non-SVR with 48 weeks of re-treatment or non-response (NR) with 24 weeks of re-treatment (*closed double circles*, Peg-IFN α-2a plus ribavirin; *closed circles* Peg-IFN α-2b plus ribavirin). *Closed double squares/closed squares*, non-SVR with 72 weeks of re-treatment (*closed double squares* Peg-IFN α-2a plus ribavirin; *closed squares*, Peg-IFN α-2b plus ribavirin)

Discussion

In the present study of the re-treatment of chronic hepatitis C patients who failed to show an SVR to Peg-IFN plus ribavirin therapy, the patients with relapse in the previous treatment showed a significant response on re-treatment compared with those with NR. This result showed similar findings to the evaluation of peg intron in control of hepatitis C chirrosis (EPIC) study of relapse and NR [10]. In addition, in the present study, p-EVR in the previous treatment was a good indicator of negative prediction for SVR on re-treatment; no patient without p-EVR in the previous treatment attained SVR on re-treatment; that is, the negative predictive value for SVR on re-treatment was 100%. Recently, genetic polymorphism near the IL28B gene has been reported to be associated with the anti-viral effect of Peg-IFN plus ribavirin combination therapy [12–15]. Among Japanese genotype 1 patients, it has been reported that those with the major single-nucleotide polymorphism (SNP) allele of IL28B (rs8099917) show an SVR rate of 39%, while those with the minor allele show an SVR rate of only 11%. Hence, in re-treatment for patients who failed to show a SVR to Peg-IFN plus ribavirin therapy, pretreatment prediction should be done by taking IL28B SNPs and the previous treatment response into account. Patients with the minor SNP allele of IL28B s who did not attain a p-EVR in the previous treatment should wait until new drugs become commercially available.

The next question is how the patients should be re-treated in order to attain an SVR on re-treatment. In the present study, the patients with a low serum HCV RNA level (less than 5 $\log_{10}$ IU/ml) at the start of re-treatment showed a significant rate of cure on re-treatment, and this is almost the same result as that previously reported [9, 10]. In the present study, one patient with NR in the previous treatment started re-treatment with HCV RNA of 52 KIU/ml and attained an RVR and SVR. HCV RNA levels declined on re-treatment among 61% (34/56) of the patients compared to the start of the previous treatment, and it is important not to miss the timing of when the HCV RNA level is low.

With respect to treatment duration among patients with HCV RNA negativity during re-treatment, 72 weeks of treatment tended to increase the SVR rate compared to

48 weeks of treatment (72 weeks, 68%, 15/22, vs. 48 weeks, 44%, 7/16, $p = 0.13$). This result was almost the same as that of the re-treatment of patients with chronic hepatitis C who do not respond to peginterferon-alpha 2b. A randomized trial (REPEAT) study [9]. Furthermore, in the present study, among the patients with relapse in the previous treatment, those who attained an SVR on re-treatment required a longer re-treatment duration than the duration of the previous treatment. In fact, the longer treatment brought about an SVR in some patients whose timing of HCV RNA negativity on re-treatment was the same as that in the previous treatment, as shown in Fig. 2. Thus, especially to be noted is that the relapsers in the previous treatment should be re-treated for a longer period than that of the previous treatment.

It has been reported that splenectomy and partial splenic embolization (PSE) are considered to make it possible for patients with cirrhosis and thrombocytopenia to initiate and continue anti-viral therapy safely, by increasing the platelet counts [16–19]. If poor adherence and inappropriate duration have contributed to a poor response in previous treatment due to thrombocytopenia, there is a possibility that increasing the platelet counts by splenectomy or PSE contributes to improving the tolerability of and adherence to re-treatment, and to increasing the SVR rate in re-treatment. In the present study, one patient with cirrhosis and thrombocytopenia who showed NR in the previous treatment owing to poor adherence to the Peg-IFN α-2b (0.78 μg/kg) regimen underwent splenectomy before re-treatment. As a result, the patient could continue with a sufficient dose of Peg-IFN (1.53 μg/kg) in the re-treatment and attained HCV negativity at re-treatment week 24 and an SVR by extended treatment. Further study is needed on the issue of the effect of splenectomy or PSE in re-treatment on the efficacy of re-treatment with Peg-IFN plus ribavirin therapy.

In the present study, the SVR rate was relatively high (56%) in patients with genotype 2. The patients who could not attain SVR on re-treatment (2 patients) had not attained a c-EVR in the previous treatment. And, among the patients with an RVR on re-treatment, all patients treated for 48 weeks attained an SVR (5 patients), while 40% (2/5) of patients treated for 24 weeks could not attain an SVR. Thus, in patients with genotype 2, as well as in those with genotype 1, the previous treatment response and response-guided therapy can be useful in decisions on the indication for re-treatment or the treatment duration on re-treatment. However, in this study, detailed analysis was not possible because of the small number of genotype 2 patients. Further investigation is needed to clarify this.

The limitation of the present study was that two types of Peg-IFN were used. As for the type of Peg-IFN, some reports have suggested that Peg-IFN α-2a has a stronger anti-viral effect than Peg-IFN α-2b [20, 21], and others have suggested that the two types of Peg-IFN have an almost equal anti-viral effect [22]. In this study, the HCV RNA negative rate at re-treatment week 12 was similar (α-2a, 59%, 13/22, vs. α-2b, 50%, 16/32, $p = 0.51$) between the patients with Peg-IFN α-2a and those with Peg-IFN α-2b. Furthermore, among 24 patients treated with Peg-IFN α-2a on re-treatment, an SVR rate of 38% was attained with 48-week treatment and an SVR rate of 60% was attained with 72-week treatment among patients with a p-EVR in the previous treatment, but no patient without a p-EVR in the previous treatment attained an SVR on re-treatment. Similarly, among 32 patients treated with Peg-IFN α-2b in re-treatment, an SVR rate of 56% was attained with 48-week treatment and an SVR rate of 79% was attained with 72-week treatment among patients with a p-EVR in the previous treatment, but no patient without a p-EVR in the previous treatment attained an SVR on re-treatment. As noted above, since the virologic responses to both Peg-IFNs among re-treated patients were similar, in this study we analyzed the effect of re-treatment without distinction of the type of Peg-IFN.

In conclusion, our results have demonstrated that the efficacy of re-treatment for genotype 1 patients who failed to show an SVR to previous treatment with Peg-IFN plus ribavirin could be predicted by the previous treatment response, especially in terms of p-EVR and a low HCV RNA level at the start of re-treatment. Re-treatment for 72 weeks led to clinical improvement for genotype 1 patients who attained HCV RNA negativity on re-treatment.

Acknowledgments Other institutions and participants in the Osaka Liver Forum are: Higashiosaka City Central Hospital, S Iio; Itami City Hospital, Y Saji; Toyonaka Municipal Hospital, M Inada; Otemae Hospital, Y Doi; Suita Municipal Hospital, T Nagase; NTT West Osaka Hospital, A Kaneko; Ashiya Municipal Hospital, T Kitada; Nishinomiya Municipal Central Hospital, H Ogawa; Saiseikai Senri Hospital, K Suzuki; Izumiotsu Municipal Hospital, S Yamagata; Osaka Kaisei Hospital, N Imaizumi; Kano General Hospital, S Kubota; Saso Hospital, M Nishiuchi; and Meiwa Hospital, Y Hayakawa. This work was supported by a Grant-in-Aid for Research on Hepatitis and BSE from the Ministry of Health, Labour and Welfare of Japan, and Scientific Research from the Ministry of Education, Science, and Culture of Japan. All authors have no financial relationships relevant to this study.

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Research Article

EASL EUROPEAN ASSOCIATION FOR THE STUDY OF THE LIVER | JOURNAL OF HEPATOLOGY

Indications and limitations for aged patients with chronic hepatitis C in pegylated interferon alfa-2b plus ribavirin combination therapy

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Background & Aims: This study investigated the efficacy and adverse effects of pegylated interferon (Peg-IFN) plus ribavirin therapy in aged patients with chronic hepatitis C (CH-C).

Methods: A total of 1040 naïve patients with CH-C (genotype 1, $n = 759$; genotype 2, $n = 281$), of whom 240 (23%) over 65 years old (y.o.), were treated with Peg-IFN alfa-2b plus ribavirin and assessed after being classified into five categories, according to age.

Results: The discontinuance rate was higher for patients over 70 y.o. (36%), the most common reason being anemia. In the presence of genotype 1, the SVR rate was similar (42–46%) among patients under 65 y.o. and declined (26–29%) among patients over 65 y.o. For patients over 65 y.o., being male (Odds ratio, OR, 3.5, $p = 0.035$) and EVR (OR, 83.3, $p < 0.001$) were significant factors for SVR, in multivariate analysis. The Peg-IFN dose was related to EVR, and when EVR was attained, 76–86% of patients over 65 y.o. achieved SVR. SVR was not achieved (0/35, 0/38, respectively) if a 1-log decrease and a 2-log decrease were not attained at week 4 and week 8, respectively. In the presence of genotype 2, the SVR rate was similar (70–71%) among patients under 70 y.o. and declined among patients over 70 y.o. (43%).

Conclusions: Aged patients up to 65 y.o. with genotype 1 and 70 y.o. with genotype 2 can be candidates for Peg-IFN plus ribavirin therapy. The response-guided therapy can be applied for aged patients with genotype 1.

Keywords: Pegylated interferon plus ribavirin therapy; Chronic hepatitis C; Aged patients.

Received 30 April 2010; received in revised form 28 July 2010; accepted 28 July 2010; available online 9 December 2010

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Abbreviations: HCV, hepatitis C virus; CH-C, chronic hepatitis C; HCC, hepatocellular carcinoma; Peg-IFN, pegylated interferon; SVR, sustained virologic response; RVR, rapid virologic response; EVR, early virologic response; LVR, late virologic response; NR, non-response; WBC, white blood cell; RBC, red blood cell; Hb, hemoglobin; Plt, platelet; G-CSF, granulocyte-macrophage colony stimulating factor.

Introduction

Pegylated interferon (Peg-IFN) plus ribavirin combination therapy has led to a marked progress in the treatment of chronic hepatitis C (CH-C) [1–4]. However, in aged patients, problems remain with respect to its anti-viral effect and tolerability [5–9]. Recently, the addition of a protease inhibitor to Peg-IFN plus ribavirin combination therapy has been reported, on the one hand, to improve the anti-viral effect, and, on the other hand, to increase side effects, especially severe anemia [10–11].

Therefore, this new therapy does not solve the problems encountered when treating aged patients.

With aging, the progression of liver fibrosis and the occurrence of hepatocellular carcinoma (HCC) have been shown to be accelerated, especially in patients over 60 y.o. [12–14]. In general, the anti-viral therapy can lead to an improvement in liver fibrosis and thus diminish the risk of HCC and ameliorate the prognosis in patients with CH-C [15–21]. Among aged patients, those results are mainly achievable upon eradication of the hepatitis C virus (HCV) [18,21]. Accordingly, the first goal of treatment of aged patients with a high-risk of HCC should be HCV elimination.

Thus, a treatment strategy, aiming at the improvement of the anti-viral efficacy in aged patients, should be established based on detailed large-scale studies.

Some points need to be further elucidated when using the Peg-IFN plus ribavirin combination therapy for the treatment of aged patients with CH-C: (i) the characteristics before treatment

that would lead to the successful elimination of HCV, (ii) the prediction factors of treatment efficacy after the initiation of the therapy, and (iii) the utility of a response-guided therapy established in the treatment.

In the present study, using a large cohort, we aimed at clarifying these points taking into account the patients' age.

Patients and methods

Patients

This study was a retrospective, multicenter trial conducted by the Osaka University Hospital and other institutions participating in the Osaka Liver Forum. A total of 1040 naïve patients with CH-C were enrolled between December 2004 and June 2007. All patients were Japanese, infected with a viral load of more than 10^5 IU/ml, and treated with a combination of Peg-IFN alfa-2b plus ribavirin. Patients were excluded from the study if they had decompensated cirrhosis or other forms of liver disease (alcohol liver disease, autoimmune hepatitis), co-infection with hepatitis B or anti-human immunodeficiency virus. This study was conducted according to the ethical guidelines of the 1975 Declaration of Helsinki and informed consent was obtained from each patient.

Treatment

All patients received Peg-IFN alfa-2b (PEGINTRON; Schering-Plough, Kenilworth, NJ, USA) plus ribavirin (REBETOL; Schering-Plough). Treatment duration was 48 weeks for patients with genotype 1 and 24 weeks for those with genotype 2. As a starting dose, Peg-IFN alfa-2b was given once weekly, at a dosage of 1.5 μg/kg, and ribavirin was given at a total dose of 600–1000 mg/day based on body weight (body weight <60 kg, 600 mg; 60–80 kg, 800 mg; >80 kg, 1000 mg), according to a standard treatment protocol for Japanese patients.

Dose reduction and discontinuance

Dose modification followed, as a rule, the manufacturer's drug information on the intensity of the hematologic adverse effects. The Peg-IFN alfa-2b dose was reduced to 50% of the assigned dose when the white blood cell (WBC) count was below 1500/mm^3, the neutrophil count below 750/mm^3 or the platelet (Plt) count below 8×10^4/mm^3, and was discontinued when the WBC count was below 1000/mm^3, the neutrophil count below 500/mm^3 or the Plt count below 5×10^4/mm^3. Ribavirin was also reduced from 1000 to 600 mg, 800 to 600 mg, or 600 to 400 mg when the hemoglobin (Hb) was below 10 g/dl, and was discontinued when the Hb was below 8.5 g/dl. Peg-IFN alfa-2b and ribavirin had to be both discontinued if there was a need to discontinue either of them. No ferric medicine or hematopoietic growth factors, such as epoetin alpha, or granulocyte–macrophage colony stimulating factor (G-CSF), were administered.

Virologic assessment and definition of virologic response

Serum HCV RNA level was quantified using the COBAS AMPLICOR HCV MONITOR test, version 2.0 (detection range 6–5000 KIU/ml; Roche Diagnostics, Branchburg, NJ) and qualitatively analyzed using the COBAS AMPLICOR HCV test, version 2.0 (lower limit of detection 50 IU/ml; Roche Diagnostics). The rapid virologic response (RVR) was defined as undetectable serum HCV RNA at week 4; the early virologic response (EVR) as undetectable serum HCV RNA at week 12; and the late virologic response (LVR) as detectable serum HCV RNA at week 12 and undetectable serum HCV RNA at week 24. Moreover, the sustained virologic response (SVR) was defined as undetectable serum HCV RNA, 24 weeks after treatment.

According to the protocol, genotype 1 patients, with less than a 2-log decrease in HCV RNA level at week 12 compared to the baseline, or with detectable serum HCV RNA at week 24, had to stop the treatment and were regarded as non-response (NR). Treatment discontinuance was evaluated except for those patients who had discontinued the treatment at up to 24 weeks, due to absence of response. Anti-viral efficacy was evaluated, for all study patients, using the intention-to-treat analysis (ITT analysis) and the per protocol analysis (PP analysis) for patients without treatment discontinuation due to side effects, and was assessed considering the definition of EVR or LVR for genotype 1, and RVR or non-RVR for genotype 2, as previously reported [1].

Assessment of drug exposure

The amounts of Peg-IFN alfa-2b and ribavirin, taken by each patient during the full treatment period, were evaluated by reviewing the medical records. The mean doses of Peg-IFN alfa-2b and ribavirin were calculated individually as averages, on the basis of the body weight at baseline: Peg-IFN alfa-2b expressed as μg/kg/week, ribavirin expressed as mg/kg/day.

Statistical analysis

Patients' baseline data are expressed as means ± SD or median values. To analyze the difference between baseline data, ANOVA or Mantel–Haenszel Chi-square test were performed. Factors associated with the viral response were assessed by univariate analysis using the Mann–Whitney *U* test or Chi-square test and multivariate analysis using logistic regression analysis. A two-tailed p value <0.05 was considered significant. The analysis was conducted with SPSS version 15.0J (SPSS Inc., Chicago, IL).

Results

Patient's profile

Baseline characteristics of the patients categorized by age are shown in Table 1.

Genotype 1 patients ($n = 759$) were distributed into five categories: 266 patients were under 55 y.o. (group 1A), 159 were 55–59 y.o. (group 1B), 149 were 60–64 y.o. (group 1C), 134 were 65–69 y.o. (group 1D), and 51 were 70 y.o. or older (group 1E). With advancing age, the male-to-female ratio and peripheral blood cell count (WBC, neutrophil count, Red blood cell (RBC), Hb, Plt) decreased significantly. Patients with a progression of liver fibrosis (METAVIR fibrosis score 3 or 4) significantly increased with age (Table 1A).

Genotype 2 patients ($n = 281$) were also distributed into five categories: 145 patients were under 55 y.o. (group 2A), 43 were 55–59 y.o. (group 2B), 38 were 60–64 y.o. (group 2C), 41 were 65–69 y.o. (group 2D), and 14 were 70 y.o. or older (group 2E). As observed in genotype 1 patients, the peripheral blood cell count decreased and the ratio of advanced fibrosis (score 3–4) increased significantly with age (Table 1B). For both genotypes, the initial doses of Peg-IFN in patients over 70 y.o. were lower than in those under 70 y.o., this was not the case for the ribavirin doses.

Dose reduction and discontinuance for adverse event

The overall discontinuance rate of treatment was 15% (140/919); 18% (112/639) for genotype 1 and 10% (28/280) for genotype 2, respectively. Table 2 shows the reason for and the rate of treatment discontinuance according to age. The discontinuance rate increased with age, being 10% (36/363) for patients under 55 y.o., 15% (27/182) for patients with 55–59 y.o., 17% (28/169) for patients with 60–64 y.o., 19% (28/147) for patients with 65–70 y.o., and significantly higher, 36%, (21/58) for patients over 70 y.o. The discontinuance of treatment due to hemolytic anemia was significantly higher for patients over 70 y.o. as compared to those under 70 y.o. (<70 y.o., 1% (9/861) vs. ⩾70 y.o., 16% (9/58), p <0.0001).

The rate without dose reduction of both drugs decreased with age (<55 y.o., 41% (171/411); 55–59 y.o., 20% (40/202); 60–64 y.o., 26% (48/187); 65–69 y.o., 23% (41/175); ⩾70 y.o., 18% (12/65)). In the presence of genotype 1, the mean dose of Peg-IFN

Table 1. Baseline characteristics of patients.

Patients with genotype 1							
Factor		<55 y.o.	55 - 59 y.o.	60 - 64 y.o.	65 - 69 y.o.	≥70 y.o.	*p* value
Number		266	159	149	134	51	
Age (y.o.)		44.4 ± 8.1	56.9 ± 1.4	62.0 ± 1.4	66.8 ± 1.4	71.4 ± 1.7	<0.001
Sex: male / female		160 / 106	64 / 95	57 / 92	54 / 80	23 / 28	<0.001
Body weight (kg)		64.6 ± 11.7	58.3 ± 9.4	58.1 ± 9.6	56.3 ± 9.3	56.3 ± 9.2	<0.001
White blood cells (/mm³)		5608 ± 1668	4901 ± 1664	4888 ± 1488	5113 ± 1426	4883 ± 1511	<0.001
Neutrophils (/mm³)		2923 ± 1214	2425 ± 1031	2559 ± 1155	2535 ± 1017	2599 ± 1149	<0.001
Red blood cells (×10⁴/mm³)		454 ± 47	432 ± 38	427 ± 40	424 ± 37	424 ± 46	<0.001
Hemoglobin (g/dl)		14.4 ± 1.5	13.8 ± 1.2	13.7 ± 1.3	13.6 ± 1.2	13.7 ± 1.4	<0.001
Platelets (×10⁴/mm³)		18.6 ± 6.2	16.3 ± 5.7	15.4 ± 5.3	15.1 ± 5.0	14.4 ± 4.2	<0.001
AST (IU/L)		62 ± 50	62 ± 45	64 ± 46	72 ± 45	64 ± 40	0.295
ALT (IU/L)		79 ± 68	76 ± 64	73 ± 63	77 ± 58	65 ± 41	0.657
Serum HCV RNA (KIU/ml)*		1800	1600	1700	1700	1700	0.691
Histology (METAVIR)†	Fibrosis, 0 - 2 / 3 - 4	177 / 19	99 / 20	90 / 19	76 / 28	21 / 9	0.001
	Activity, 0 - 1 / 2 - 3	117 / 79	63 / 56	59 / 50	47 / 57	13 / 16	0.146
Peg-IFN dose (μg/kg/week)¶		1.47 ± 0.14	1.47 ± 0.16	1.46 ± 0.18	1.44 ± 0.18	1.36 ± 0.24	<0.001
Ribavirin dose (mg/kg/day)¶		11.5 ± 1.1	11.5 ± 1.4	11.5 ± 1.4	11.5 ± 1.7	11.2 ± 2.2	0.65

Patients with genotype 2							
Factor		<55 y.o.	55 - 59 y.o.	60 - 64 y.o.	65 - 69 y.o.	≥70 y.o.	*p* value
Number		145	43	38	41	14	
Age (y.o.)		40.9 ± 8.9	56.7 ± 1.3	62.3 ± 1.4	66.7 ± 1.5	71.8 ± 1.8	<0.001
Sex: male / female		78 / 67	17 / 26	17 / 21	18 / 23	6 / 8	0.441
Body weight (kg)		63.4 ± 12.0	59.5 ± 11.5	58.6 ± 11.7	58.5 ± 9.8	55.9 ± 6.8	0.783
White blood cells (/mm³)		6011 ± 1965	4874 ± 1346	4982 ± 1210	5079 ± 1877	4414 ± 871	<0.001
Neutrophils (/mm³)		3214 ± 1511	2468 ± 971	2576 ± 950	2492 ± 1119	2521 ± 683	0.001
Red blood cells (×10⁴/mm³)		454 ± 48	430 ± 42	432 ± 50	430 ± 43	408 ± 48	<0.001
Hemoglobin (g/dl)		14.3 ± 1.6	13.5 ± 1.3	13.9 ± 1.4	13.9 ± 1.3	13.3 ± 1.2	0.001
Platelets (×10⁴/mm³)		21.3 ± 5.4	18.3 ± 6.1	17.0 ± 5.2	15.8 ± 5.4	13.9 ± 4.7	<0.001
AST (IU/L)		53 ± 59	57 ± 45	55 ± 38	83 ± 48	68 ± 29	0.029
ALT (IU/L)		65 ± 59	73 ± 70	68 ± 62	105 ± 62	78 ± 43	0.008
Serum HCV RNA (KIU/ml)*		1700	1100	900	1100	500	0.008
Histology (METAVIR)‡	Fibrosis, 0 - 2 / 3 - 4	102 / 0	25 / 3	29 / 2	21 / 9	7 / 1	<0.001
	Activity, 0 - 1 / 2 - 3	68 / 34	18 / 10	18 / 13	9 / 21	5 / 3	0.01
Peg-IFN dose (μg/kg/week)¶		1.48 ± 0.16	1.48 ± 0.14	1.45 ± 0.18	1.46 ± 0.15	1.28 ± 0.26	0.001
Ribavirin dose (mg/kg/day)¶		11.5 ± 1.1	11.4 ± 1.2	11.5 ± 1.4	11.3 ± 1.6	11.0 ± 1.4	0.55

*, Data shown are median values.
†, 201 Missing.
‡, 82 Missing.
¶, Initial doses.

during the whole treatment period was lower (1.1 ± 0.3 μg/kg/week) for patients over 70 y.o. than for those under 70 y.o. (1.3 ± 0.3 μg/kg/week) and that of ribavirin decreased with age (<55 y.o., 10.3 ± 1.9 mg/kg/day; 55–59 y.o., 9.8 ± 1.9 mg/kg/day; 60–64 y.o., 9.3 ± 2.3 mg/kg/day; 65–69 y.o., 9.2 ± 2.3 mg/kg/day; ≥70 y.o., 8.5 ± 2.5 mg/kg/day). The same tendency was observed with genotype 2.

Sustained virologic response

In genotype 1 patients, the overall SVR rate was 40% (305/759), being 46% (123/266) for group 1A, 44% (70/159) for group 1B, 42% (62/149) for group 1C, 26% (35/134) for group 1D, and 29% (15/51) for group 1E, following ITT analysis. The same tendency was observed using the PP analysis ($n = 647$). The SVR rates for patients over 65 y.o. were significantly lower than those for patients under 65 y.o. (ITT analysis: ≥65 y.o., 27% vs. <65 y.o., 44%, $p < 0.0001$; PP analysis: ≥65 y.o., 31% vs. <65 y.o., 50%, $p < 0.0001$) (Fig. 1A). Among genotype 1 patients over 65 y.o., the SVR rate was significantly lower for female patients than for male patients (ITT analysis: male, 40% (31/77) vs. female, 18% (19/108), $p < 0.001$; PP analysis: male, 49% (27/55) vs. female, 20% (18/90), $p < 0.001$).

Moreover, for genotype 2 patients, the overall SVR rate was 78% (220/281), being 88% (128/145) for group 2A, 70% (30/43) for group 2B, 71% (27/38) for group 2C, 71% (29/41) for group 2D, and 43% (6/14) for group 2E, following ITT analysis. The same tendency was observed with the PP analysis ($n = 253$). The SVR rates for patients over 70 y.o. were significantly lower than those for patients under 70 y.o. (ITT analysis: ≥70 y.o., 43% vs. <70 y.o., 80%, $p < 0.0001$; PP analysis: ≥70 y.o., 56% vs. <70 y.o., 85%, $p < 0.05$) (Fig. 1B). Among patients over 70 y.o. with genotype 2, the difference according to gender was not clear because of the small sample.

Table 2. Reasons for treatment discontinuation.

Factor	<55 y.o. (n = 363)	55 - 59 y.o. (n = 182)	60 - 64 y.o. (n = 169)	65 - 69 y.o. (n = 147)	≥70 y.o. (n = 58)	Total (n = 919)
Neutropenia	2	3	0	0	0	5
Thrombopenia	1	0	1	1	0	3
Anemia	0	4	3	2	9	18
Fatigue	1	1	3	3	1	9
Gastrointestinal disorder	2	1	0	0	1	4
Cough, Dyspnea	1	0	3	0	0	4
Vertigo	1	0	0	0	3	4
Psychosis (depression)	7 (3)	7 (3)	4 (4)	3 (3)	2 (2)	23
Rash	5	2	5	7	1	20
Thyroid dysfunction	2	0	2	0	0	4
Fundal hemorrhage	0	2	0	2	0	4
Drug-induced hepatitis	3	1	0	0	0	4
Interstitial pneumonia	0	1	0	1	1	3
Cerebral hemorrhage, infarction	2	0	0	1	0	3
Others	9	5	7	8	3	32
Total	36 (10%)	27 (15%)	28 (17%)	28 (19%)	21 (36%)	140 (15%)

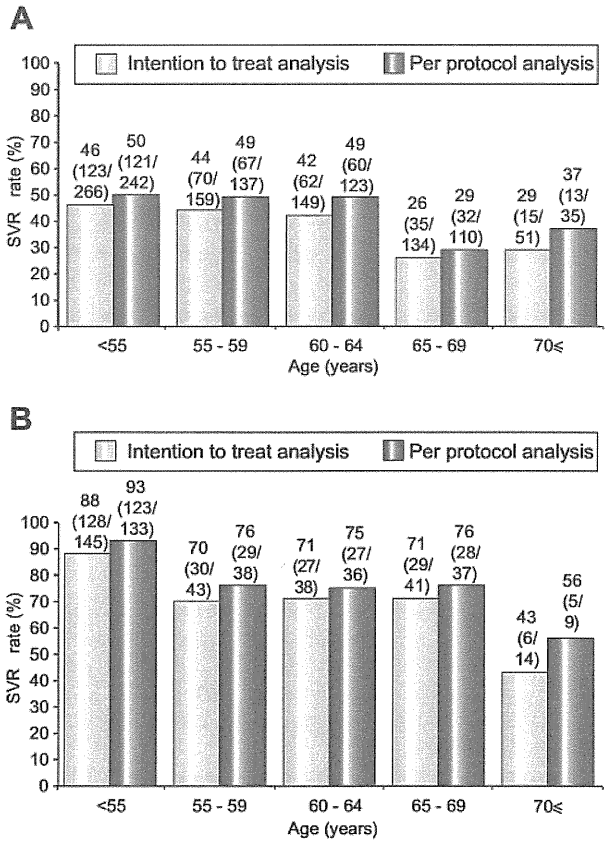

Fig. 1. SVR rate according to age. (A) Genotype 1. (B) Genotype 2.

Timing of HCV RNA negativation for genotype 1, according to age

Treatment responses distributing EVR, LVR, and NR according to age are shown in Fig. 2. The rates of NR were similar in patient groups under 65 y.o. (30–36%), but increased in almost half of

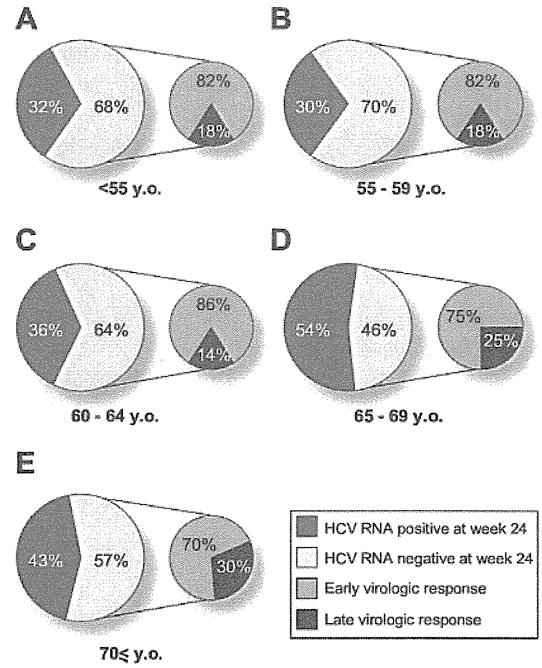

Fig. 2. Antiviral effect during treatment according to age. (A) <55 y.o. (B) 55–59 y.o. (C) 60–64 y.o. (D) 65–69 y.o. (E) ≥70 y.o.

the patients over 65 y.o. (p <0.0001). Moreover, among the virologic responders, the proportion of LVR tended to increase in patients over 65 y.o. (25–30%) compared to patients under 65 y.o. (14–18%) (p = 0.06).

SVR rate according to the timing of HCV RNA negativation

SVR rates according to EVR or LVR in genotype 1, and RVR or non-RVR in genotype 2 are summarized in Table 3. Genotype 1 patients with EVR achieved high SVR rates regardless of age; in particular, if EVR had been attained, 76% of patients with 65–69

Table 3. SVR rate according to genotype and viral response in patients responding to PEG-IFN plus ribavirin combination therapy.

Factor	<55 y.o.	55 - 59 y.o.	60 - 64 y.o.	65 - 69 y.o.	≥70 y.o.
Genotype 1					
with EVR, % (n)	85 (114/134)	79 (62/79)	81 (55/68)	76 (29/38)	86 (12/14)
with LVR, % (n)	23 (7/30)	29 (5/17)	46 (5/11)	23 (3/13)	17 (1/6)
Genotype 2					
with RVR, % (n)	93 (57/61)	82 (14/17)	85 (17/20)	92 (11/12)	100 (4/4)
without RVR*, % (n)	96 (22/23)	60 (6/10)	57 (4/7)	50 (4/8)	0 (0/3)

RVR, rapid virologic response.
EVR, early virologic response.
LVR, late virologic response.
*, Serum HCV RNA was detectable at week 4, but undetectable at week 24.

Table 4. Multivariate analysis for the factors associated with SVR among all patients.

Factor	Category	Odds ratio	95% CI	p
Age (y.o.)	<65 / ≥65	0.485	0.295 - 0.799	0.005
Sex	male / female	0.524	0.353 - 0.777	0.001
Platelets ($\times 10^4$/mm^3)	<12 / ≥12	1.780	1.039 - 3.049	0.040
Serum HCV RNA (KIU/ml)	<2000 / ≥2000	0.599	0.401 - 0.896	0.010
Histology (METAVIR): Fibrosis	0 - 2 / 3 - 4	0.599	0.333 - 1.076	0.090

y.o. and 86% of patients over 70 y.o. achieved SVR, and these SVR rates compared favorably with those of younger patients. On the other hand, the SVR rates for patients with LVR ranged from 17% to 46%, which were lower than those for EVR patients in each age group, and no significant differences of SVR rates were found among LVR patients by age.

With genotype 2, patients with RVR achieved high SVR rates ranging from 82% to 100% regardless of age. Even for patients without RVR, 96% of those under 55 y.o. attained SVR, a rate that was significantly higher than that for patients over 55 y.o. (50%, 14/28) (p <0.001).

Factors associated with SVR for genotype 1

The factors associated with SVR were assessed for the variables shown in Table 1. The factors selected as significant by the univariate analysis: age, gender, WBC, neutrophils, RBC, Hb, Plt, aspartate aminotransferase, serum HCV RNA level, the degree of liver fibrosis, and the initial dose of Peg-IFN, were evaluated by multivariate logistic regression analysis. The factor of age over 65 y.o. was the independent factor for SVR (p = 0.005), apart from the gender (p = 0.001), Plt value (p <0.05), and serum HCV RNA level (p = 0.01) (Table 4).

Factors associated with EVR and SVR for patients over 65 y.o. with genotype 1

The results of univariate analysis for EVR among patients over 65 y.o. are shown in Table 5A. Gender, Plt value, and mean dose of Peg-IFN during the first 12 weeks were factors significantly associated with EVR. In multivariate analysis, the mean dose of Peg-IFN during the first 12 weeks was the independent factor for EVR (p = 0.03), apart from gender (p = 0.002) (Table 5B). The EVR rates were 41% (41/101) in patients who received ⩾1.2 μg/kg/week on average during the first 12 weeks, and declined to 36% (8/22) in patients given 0.9–1.2 μg/kg/week of Peg-IFN, and to 14% (3/22) in patients administered with <0.9 μg/kg/week of Peg-IFN.

The baseline and on-treatment factors, which are correlated with the SVR among the patients over 65 y.o., were assessed by univariate and multivariate analyses. Univariate analysis showed that factors significantly associated with SVR were gender and virologic response (Table 6A), and they were also selected as significant independent factors in multivariate analysis (p = 0.035, p <0.001) (Table 6B).

Negative prediction of SVR for patients over 65 y.o. with genotype 1

We tried positive and negative predictions of SVR for aged patients, focusing on the decrease of HCV RNA at treatment week 4 and 8. The SVR rate was 47% (29/62) for patients with more than a 1-log decrease in HCV RNA level at week 4, while no patients with less than a 1-log decrease at week 4 attained SVR (0/35) (p <0.0001). Similarly, 55% (35/64) of patients with more than a 2-log decrease at week 8 attained SVR, whereas no patients with less than a 2-log decrease at week 8 attained SVR (0/38) (p <0.0001).

Discussion

Peg-IFN plus ribavirin combination therapy can improve anti-viral efficacy and is presently recommended as first-line therapy [1–4]. However, with respect to aged patients with CH-C, there have been only a few small-scale cohort studies which reported poor anti-viral effect and poor tolerability in comparison with non-aged patients [5–9]. The problem in the treatment of aged patients with CH-C is most serious in Japan, because HCV carriers in Japan are 10–20 years older than those in the United States and European countries [22]. Therefore, in the present study, we examined the efficacy and prevalence of side effects with a focus on patient's age using a large-scale cohort.