≥65 years) showed increases in reported adverse events and laboratory abnormalities in older patients, but these differences did not present a barrier to treatment as no premature discontinuation of study treatment occurred in any patient. Analysis of safety data according to the presence or absence of cirrhosis did not indicate clinically important differences in safety or tolerability of the 12-week sofosbuvir plus ribavirin regimen.

Consistent with previous reports, the results of this study confirm the high barrier to resistance afforded by the sofosbuvir plus RBV treatment regimen. Rapid viral suppression was observed with all patients achieving HCV RNA undetectable status by week 4, with no virologic breakthrough observed during treatment in any of the 153 patients. The percentage of patients who relapsed after treatment was low (3%), and none of the subjects who relapsed had S282T or other nucleoside inhibitor resistance-associated variants. No change in susceptibility to sofosbuvir or ribavirin compared with the corresponding baseline or wild-type reference was observed at the relapse time point.

The main limitation of this study was the lack of a control arm to allow direct comparison with interferon-based regimens. Several considerations guided our choice of an uncontrolled study design. Adding an interferon-based control arm would have required exclusion of patients who were ineligible to receive or intolerant of interferon – an important and substantial proportion of patients – as well as previously treated patients, for whom further interferon treatment is not an option. Moreover, given that Peg-IFNα is administered by subcutaneous injection, blinding of treatment arms would not have been possible.

In conclusion, treatment with the all-oral, interferon-free combination of sofosbuvir and RBV resulted in high rates of sustained virologic response in both treatment-naïve and previously treated Japanese patients with chronic genotype 2 HCV infection. The degree of antiviral efficacy coupled with a favourable safety and tolerability profile, including patients with cirrhosis and those aged 65 and older, suggest that this combination may fill an important unmet medical need in Japan.

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SUPPORTING INFORMATION

Additional Supporting Information may be found in the online version of this article:

Fig. S1. Patient disposition.

Table S1. Reasons for screen failure.

Table S2. Calculation of the adjusted historical control rate.

Table S3. SVR12 by subgroup.

Table S4. Characteristics of patients who relapsed.

Table S5. Common adverse events by age group.

Table S6. Common adverse events by cirrhosis status.

Table S7. Gilead sciences grading scale for severity of adverse events and laboratory abnormalities.

Hepatology Research

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Original Article

Simeprevir (TMC435) once daily with peginterferon-α-2b and ribavirin in patients with genotype 1 hepatitis C virus infection: The CONCERTO-4 study

Hiromitsu Kumada,[1] Norio Hayashi,[2] Namiki Izumi,[3] Takeshi Okanoue,[4] Hirohito Tsubouchi,[5] Hiroshi Yatsuhashi,[6] Mai Kato,[7] Ki Rito,[7] Yuji Komada,[8] Chiharu Seto[7] and Shoichiro Goto[7]

[1]Toranomon Hospital, [3]Musashino Red Cross Hospital, [7]Research and Development, Clinical Science Division, [8]Research and Development, Japan Clinical Operations Division, Janssen Pharmaceutical K.K., Tokyo, [2]Kansai-Rosai Hospital, Hyogo, [4]Saiseikai Suita Hospital, Osaka, [5]Kagoshima University Medical and Dental Hospital, Kagoshima, and [6]National Hospital Organization Nagasaki Medical Center, Nagasaki, Japan

Aim: The efficacy and safety of simeprevir in combination with peginterferon-α-2b and ribavirin (PEG IFN-α-2b/RBV) were investigated in patients infected with hepatitis C virus (HCV) genotype 1 who were treatment-naïve or had previously received interferon (IFN)-based therapy.

Methods: CONCERTO-4 (NCT01366638) was an open-label, non-comparative, multicenter study of once-daily simeprevir (TMC435) 100 mg in combination with PEG IFN-α-2b/RBV in treatment-naïve and -experienced patients (prior relapsers or non-responders to IFN-based therapy) with chronic HCV genotype 1 infection. Twelve-week combination treatment was followed by 24/48-week response-guided PEG IFN-α-2b/RBV therapy for treatment-naïve patients and prior relapsers, and 48-week PEG IFN-α-2b/RBV therapy for prior non-responders. Patients were followed for 72 weeks after treatment initiation. The proportions of patients with sustained viral response (SVR; undetectable HCV RNA) at treatment end and 12 weeks after the last treatment (SVR12) were among the major efficacy end-points. Safety, including adverse events (AE), was monitored.

Results: Of the 79 patients treated, the proportion achieving SVR12 was highest among treatment-naïve patients (91.7%) and prior relapsers (100%) versus 38.5% of prior non-responders. All treatment-naïve patients and prior non-responders who achieved SVR12 also achieved SVR at treatment end and 24 weeks after last dose; 96.6% of prior relapsers achieved both end-points. Most AE were of grade 1 or 2 severity. Grade 3 AE occurred in 17 patients, most frequently neutropenia (6.3%).

Conclusion: Simeprevir combined with PEG IFN-α-2b/RBV was effective in patients infected with HCV genotype 1, both for initial treatment of naïve patients and for retreatment of patients in whom previous IFN-based therapy had failed.

Key words: chronic hepatitis C, direct-acting antiviral, protease inhibitor, simeprevir (TMC435), sustained virologic response

Correspondence: Dr Shoichiro Goto, Research and Development, Clinical Science Division, Janssen Pharmaceutical K.K., 5-2 Nishi-kanda 3-chome, Chiyoda-ku, Tokyo 101-0065, Japan. Email: sgoto2@its.jnj.com

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Author contribution: Dr Kumada, Dr Yatsuhashi, Dr Okanoue, Dr Tsubouchi, Dr Hayashi and Dr Izumi: conception and interpretation of data, revising the draft for critically important intellectual content and final approval of the draft to be published. Kato, Rito, Komada, Seto and Goto: design, analysis and interpretation of data, drafting and revising the draft and final approval of the draft to be published.

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INTRODUCTION

APPROXIMATELY 1.5–2 MILLION Japanese people are infected with hepatitis C virus (HCV), with the majority being infected with HCV genotype 1b.[1] Chronic infection with HCV is a major cause of liver disease,[2] and is estimated to account for more than 70% of hepatocellular carcinoma cases, one of the most common causes of cancer death in Japan.[3]

Combination therapy with weekly peginterferon-α (PEG IFN-α) injections and twice-daily oral ribavirin (RBV) for 48–72 weeks has been standard care for HCV genotype 1 infection for many years.[4–6] However, treatment discontinuations and dose reductions are common owing to the wide range of adverse events (AE) associated with PEG IFN-α/RBV therapy, including influenza-like symptoms, anemia and depression.[7–9]

Novel direct-acting antiviral agents, including protease inhibitors (PI) that target the HCV NS3/4A serine protease, have recently become available and are recommended for use in combination with PEG IFN-α/RBV.[6] The addition of PI to PEG IFN-α/RBV has improved treatment outcomes substantially in both treatment-naïve and treatment-experienced patients.[6,10–16] Sustained virologic response (SVR) rates of 60–88% have been reported for the first-generation PI in combination with PEG IFN-α/RBV in untreated and previously treated relapsed HCV infection,[10–16] compared with rates of 40–50% with PEG IFN-α/RBV alone.[7–9,17,18] This has enabled the use of shorter courses of PEG IFN-α/RBV than the standard 48 weeks.[13,19] However, currently available PI in combination with PEG IFN-α/RBV are associated with higher incidences of anemia, dysgeusia, rash and nausea than PEG IFN-α/RBV alone,[10–13,15,16] and high rates of patient discontinuation.[11,14] In addition, currently available PI require multiple daily dosing. Patients infected with HCV would benefit from novel agents with improved tolerability and more convenient dosing schedules.

Simeprevir (TMC435) is a once-daily, oral HCV NS3/4A PI, with potent antiviral activity against HCV genotype 1,[20] as well as against isolates of genotypes 2 and 4–6.[21] Simeprevir combined with PEG IFN-α-2a/RBV has demonstrated good tolerability and high SVR rates in both treatment-naïve and treatment-experienced patients infected with HCV genotype 1 in international studies[22–24] and in phase III studies in Japan (CONCERTO-1,-2 and -3).[25,26]

We report the results of a phase III, open-label, non-comparative study (CONCERTO-4) conducted in Japan to investigate the efficacy and safety of simeprevir in combination with PEG IFN-α-2b/RBV in patients infected with HCV genotype 1 who were treatment-naïve or had previously received interferon (IFN)-based therapy.

METHODS

Patients

ELIGIBLE PATIENTS WERE aged 20–70 years with chronic HCV genotype 1 infection and plasma HCV RNA of 5.0 $\log_{10}$ IU/mL or more at screening. Treatment-naïve patients must not have received prior treatment with any approved or investigational HCV drug (including IFN). Patients who had previously received IFN-based therapy for 24 weeks or more were eligible provided their last treatment was administered 60 days or more before the study start. Treatment-experienced patients were classified as prior relapsers (i.e. patients who had undetectable levels of HCV RNA at the last assessment while on IFN-based therapy and subsequent detectable levels of HCV RNA within 12 months from their last treatment), or prior non-responders (i.e. patients who did not achieve undetectable HCV RNA on prior IFN-based therapy or who had discontinued IFN-based therapy within 24 weeks of treatment initiation due to <2 $\log_{10}$ IU/mL reduction from baseline in HCV RNA at week 12 of treatment). All patients provided written informed consent.

Exclusion criteria included liver cirrhosis or hepatic failure, liver disease of non-HCV etiology, infection/co-infection with non-genotype 1 HCV, hepatitis B virus, or HIV-1 or HIV-2, any condition that required caution with PEG IFN-α-2b or RBV therapy, and any other clinically significant disease, organ transplant or defined laboratory abnormalities at screening. In addition, treatment-experienced patients were not eligible if they had received treatment with any HCV therapy other than IFN, PEG IFN or RBV, or if they had discontinued previous therapy due to an AE considered likely to be treatment-limiting during PEG IFN-α-2b/RBV therapy.

Study design

This was an open-label, non-comparative, multicenter study to assess the efficacy and safety of simeprevir (TMC435) combined with PEG IFN-α-2b/RBV in treatment-naïve and treatment-experienced (prior relapsers or non-responders to IFN-based therapy) patients with chronic HCV genotype 1 (NCT01366638). The study was conducted at 14 sites in Japan from 1 April 2011 to 20 November 2012. The study was

approved by the relevant institutional review boards and was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice Guidelines.

Eligible patients received oral simeprevir 100 mg once daily (Q.D.) plus PEG IFN-α-2b/RBV for 12 weeks. In treatment-naïve patients and prior relapsers, this was followed by response-guided therapy (RGT) with PEG IFN-α-2b/RBV until week 24 or 48. Treatment-naïve patients and prior relapsers who achieved HCV RNA of less than 1.2 $\log_{10}$ IU/mL detectable or undetectable levels at week 4, with undetectable levels at week 12, stopped PEG IFN-α-2b/RBV therapy at week 24, while all others continued to week 48. All prior non-responders received PEG IFN-α-2b/RBV until week 48. All patients were followed for 72 weeks after treatment initiation.

Patients had to discontinue simeprevir but could continue with PEG IFN-α-2b/RBV if they experienced grade 4 elevations of aspartate aminotransferase (AST) or alanine aminotransferase (ALT) and the value was more than 2 times baseline, or if they experienced grade 4 blood bilirubin elevations and bilirubin values were the same or higher at retesting. All study medications were stopped if patients experienced grade 4 AE or laboratory abnormalities that were not considered to be related to simeprevir specifically or were not expected toxicities of PEG IFN-α-2b/RBV or HCV infection, or if patients experienced grade 3/4 skin events/allergic reactions, or worsening of hepatic disease. Additionally, all study medications were stopped if the following predefined virologic stopping criteria were met: less than 2 $\log_{10}$ IU/mL reduction in HCV RNA at week 12 relative to baseline (treatment-naïve patients and prior relapsers); HCV RNA levels of more than 2 $\log_{10}$ IU/mL at week 12 (prior non-responders); and confirmed detectable HCV RNA of 1.2 $\log_{10}$ IU/mL or more at weeks 24 or 36 (all patients). Patients who discontinued therapy proceeded immediately into follow-up.

The major efficacy end-point was the proportion of patients with undetectable HCV RNA at the end of treatment and 12 weeks after the last treatment (SVR12). Other efficacy end-points included the proportion of patients with: undetectable HCV RNA at end of treatment and 24 weeks after the last treatment (SVR24); undetectable HCV RNA at end of treatment and 4 weeks after the last treatment (SVR4); rapid virologic response (RVR; undetectable HCV RNA at week 4); complete early virologic response (cEVR; undetectable HCV RNA at week 12); undetectable HCV RNA at the end of treatment; viral breakthrough (increase of >1 $\log_{10}$ IU/mL in plasma HCV RNA level from the lowest level reached or plasma HCV RNA level >2.0 $\log_{10}$ IU/mL in patients whose plasma HCV RNA level had previously been <1.2 $\log_{10}$ IU/mL detectable or undetectable); viral relapse (detectable or quantifiable plasma HCV RNA during the post-treatment follow-up period in patients who had undetectable plasma HCV RNA at the end of treatment); and normalization of ALT. Tolerability and safety (AE, clinical laboratory parameters and vital signs) were secondary end-points.

Treatment administration

Simeprevir 100 mg was administered orally Q.D. as a single capsule. No simeprevir dose adjustments were permitted but, at the investigator's discretion, dosing could be interrupted for 4 days or less due to AE. PEG IFN-α-2b (PegIntron®; Merck Sharp & Dohme, Whitehouse Station, NJ, USA) was administered weekly as an s.c. injection (1.5 μg/kg body weight), and RBV (Rebetol®, Merck Sharp & Dohme) was administered as oral capsules (600–1000 mg total daily dose, according to body weight). Dose change, temporary interruption or discontinuation of PEG IFN-α-2b and RBV had to be conducted in accordance with the manufacturer's prescribing information. Patients were hospitalized for at least 1 week, starting on the first day of treatment. Use of erythropoiesis-stimulating agents and medications acting on the immune system was not permitted during treatment.

Study assessments

Plasma HCV RNA was quantified at screening, at baseline, on day 3, and at weeks 1, 2, 3, 4, 8, 12, 16, 20 and 24 (all patients), and weeks 28, 36, 48, 60 and 72 (patients receiving PEG IFN-α-2b/RBV until week 24), or weeks 28, 36, 42, 48, 52, 60 and 72 (patients receiving PEG IFN-α-2b/RBV until week 48). Levels were determined at a central laboratory using Roche COBAS® TaqMan® HCV Auto (Roche Molecular Diagnostics, Pleasanton, CA, USA) with a lower limit of quantification of 1.2 $\log_{10}$ IU/mL.

Sequence analysis of the HCV NS3 protease domain was performed at baseline and in patients with simeprevir treatment failure (viral breakthrough, meeting virologic stopping rule, detectable HCV RNA at end of treatment or viral relapse). The analysis of baseline polymorphisms focused on detecting previously characterized HCV genotype 1 amino acid substitutions in the NS3 region at positions 36, 43, 54, 80, 122, 138, 155, 156, 168 and 170 that have been associated with reduced susceptibility to simeprevir and other HCV NS3 PI *in vitro*.[27,28]

Safety and tolerability were evaluated throughout the entire treatment period, from first study medication intake until 28 days after the last dose. Severity of AE was graded by investigators according to the World Health Organization (WHO) grading scale. Vital sign monitoring, electrocardiograms, physical examinations and clinical laboratory tests were performed at regular intervals during the study period. Severity of laboratory abnormalities was classified according to the WHO grading scale.

Statistical analysis

A sample size of 70 patients was deemed sufficient to give a 97% probability of detecting an AE of special interest with 5% or more incidence.

Efficacy analyses were performed on the full analysis set (i.e. all patients who received the study medication and had post-baseline efficacy assessment data). The safety population included all patients who received at least one dose of simeprevir.

Ninety-five percent confidence intervals (CI) around the SVR12, SVR24 and SVR4 rates were calculated for each group. Descriptive statistics and tabulation were used to summarize baseline characteristics. All statistical analyses were performed using SAS® version 9.2 (SAS Institute, Cary, NC, USA).

RESULTS

Patients

IN TOTAL, 97 patients were screened and 79 received treatment (24 treatment-naïve patients, 29 prior relapsers and 26 prior non-responders) (Fig. 1). All 79 patients who received treatment were included in the full analysis set and safety populations. All study medications were completed by 65 patients (82.3%). The rate of treatment completion was lowest among prior non-responders (57.7% vs 91.7% for treatment-naïve patients and 96.6% for prior relapsers). Of the 14 patients who discontinued medications, one patient discontinued simeprevir and subsequently discontinued PEG IFN-α-2b/RBV, nine patients discontinued PEG IFN-α-2b/RBV after completing simeprevir treatment and four patients discontinued all study medications at the same time. The main reason for treatment discon-

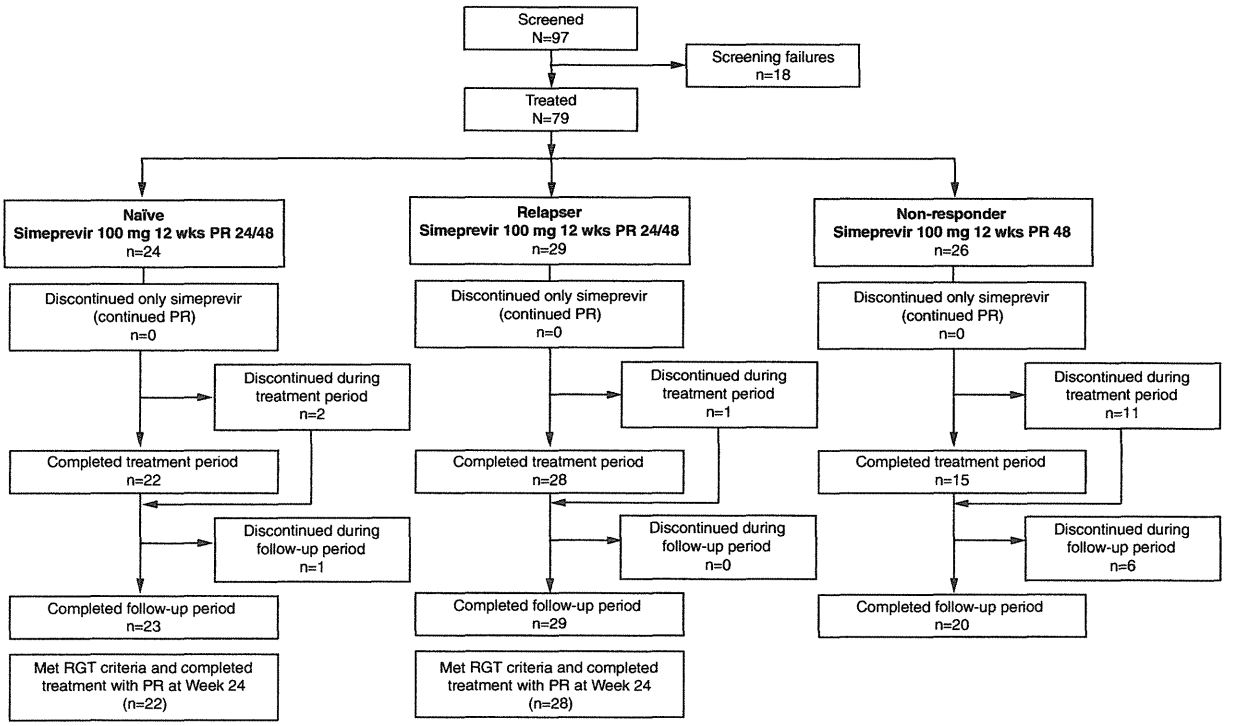

Figure 1 Patient disposition. PR, peginterferon-α-2b and ribavirin; RGT, response-guided treatment; wks, weeks.

Table 1 Patient demographics and baseline characteristics (full analysis set)

Characteristic	Treatment-naïve ($n = 24$)	Prior relapsers ($n = 29$)	Prior non-responders ($n = 26$)
Male, *n* (%)	8 (33.3)	16 (55.2)	13 (50.0)
Age, years, median (range)	60 (37–68)	60 (38–70)	53 (45–69)
Age <65 years, *n* (%)	19 (79.2)	20 (69.0)	22 (84.6)
BMI, kg/m^2, median (range)	22.95 (18.1–30.2)	22.5 (18.1–31.9)	22.4 (16.9–34.3)
IL28B genotype (SNP rs8099917)			
TT, n (%)	16 (66.7)	26 (89.7)	2 (7.7)
TG/GG, *n* (%)	8 (33.3)	3 (10.3)	24 (92.3)
IL28B genotype (SNP rs12979860)			
CC, *n* (%)	16 (66.7)	26 (89.7)	2 (7.7)
CT/TT, *n* (%)	8 (33.3)	3 (10.3)	24 (92.3)
Genotype 1b, *n* (%)	24 (100.0)	29 (100.0)	25 (96.2)
Baseline HCV RNA, $\log_{10}$ IU/mL, median (range)	6.6 (5.4–7.0)	6.6 (4.9–7.4)	6.5 (5.1–7.4)
METAVIR score, category, *n* (%)†	$n = 6$	$n = 6$	$n = 7$
F0	0	0	0
F1	5 (83.3)	4 (66.7)	5 (71.4)
F2	1 (16.7)	1 (16.7)	2 (28.6)
F3	0	1 (16.7)	0
F4	0	0	0
Platelets ($\times 10^9$/L), *n* (%)			
<150	5 (20.8)	9 (31.0)	11 (42.3)
≥150	19 (79.2)	20 (69.0)	15 (57.7)
Prior therapy, *n* (%)			
IFN only	N/A	1 (3.4)	0
IFN/RBV	N/A	0	3 (11.5)
PEG IFN only	N/A	0	0
PEG IFN/RBV	N/A	28 (96.6)	23 (88.5)
ALT			
<50 IU/mL	16 (66.7)	20 (69.0)	13 (50.0)
≥50 IU/mL	8 (33.3)	9 (31.0)	13 (50.0)
Total bilirubin (mg/dL), median (range)	0.7 (0.3–1.8)	0.8 (0.4–2.2)	0.8 (0.3–1.1)
Hemoglobin (g/dL), median (range)	14.2 (12.4–16.3)	14.4 (11.5–17.0)	13.9 (12.2–16.6)
Neutrophils ($\times 10^2$/μL), median (range)	25.4 (12.1–51.2)	25.4 (10.1–48.1)	22.2 (9.6–35.8)
Platelets ($\times 10^4$/μL), median (range)	17.1 (12.2–27.5)	16.3 (9.6–33.3)	15.4 (11.0–20.5)

†Available for patients who had a liver biopsy within two years prior to informed consent or during the screening period.
ALT, alanine aminotransferase; BMI, body mass index; HCV, hepatitis C virus; IFN, interferon; N/A, not applicable; PEG IFN, peginterferon; RBV, ribavirin; SNP, single nucleotide polymorphism.

tinuation was meeting the virologic stopping criteria (eight patients, all prior non-responders).

Demographic and disease characteristics at baseline were generally comparable across the three patient groups (Table 1), with a few notable exceptions for sex, *IL28B* genotype and baseline platelet counts. Median age was 60 years (range, 37–70), with 22.8% of patients aged 65 years or more. Most treatment-naïve patients and prior relapsers had major allele TT and CC genotypes for *IL28B* rs9088817 and rs12979860 polymorphisms, respectively. By contrast, most prior non-responders had minor alleles TG/GG and CT/TT at these loci. All but one patient (prior non-responder) had HCV genotype 1b; median HCV RNA at baseline was 6.5 $\log_{10}$ IU/mL. Most prior relapsers and prior non-responders had previously been treated with PEG IFN plus RBV. Platelet counts at baseline were slightly lower in prior non-responders, with 42.3% having counts of less than 150×10^9/L versus 31.0% or less of patients in the other groups.

Efficacy

SVR

The proportion of patients achieving SVR4, SVR12 (major efficacy end-point), and SVR24 is shown in Fig. 2. The proportion of patients achieving SVR12 was

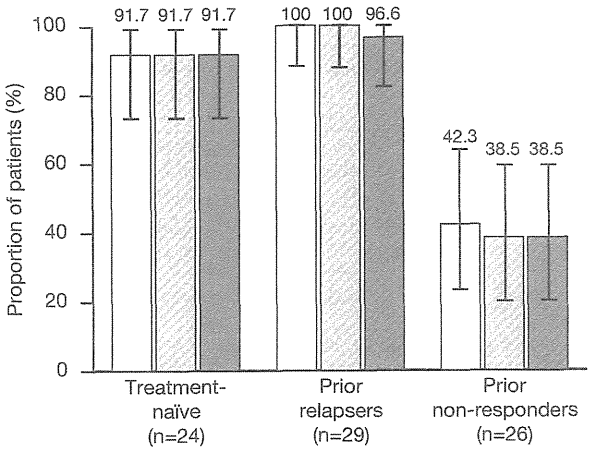

Figure 2 Sustained virologic response at 4, 12 (major end-point) and 24 weeks after the end of treatment. Bars indicate 95% confidence intervals. †$n = 12$. SVR, sustained virologic response (undetectable hepatitis C virus RNA); SVR4, SVR at end of actual treatment and at 4 weeks after the last treatment; SVR12, SVR at end of actual treatment and at 12 weeks after the last treatment (major efficacy end-point); SVR24, SVR at end of actual treatment and at 24 weeks after the last treatment. □, SVR4; ▫, SVR12; ■, SVR24.

highest among treatment-naïve patients (91.7%; 95% CI, 73.0–99.0%) and prior relapsers (100%; 95% CI, 88.1–100%). Only two patients in the treatment-naïve group did not achieve SVR12; both had undetectable HCV RNA at end of treatment, but had viral relapse at the SVR4 time point. Among prior non-responders, 10 patients (38.5%; 95% CI, 20.2–59.4%) achieved SVR12, 11 patients had detectable HCV RNA at end of treatment, four patients had viral relapse at the SVR4 time point, and one patient had discontinued follow-up before the SVR12 time point.

The SVR24 rate was 91.7% (95% CI, 73.0–99.0%) among treatment-naïve patients and 96.6% (95% CI, 82.2–99.9%) for prior relapsers (Fig. 2). All treatment-naïve patients and prior non-responders who achieved SVR12 also achieved SVR24, while 28 of 29 prior relapsers achieved both end-points (one patient experienced viral relapse at week 24 of follow-up).

Twenty-two (91.7%) treatment-naïve patients and 28 (96.6%) prior relapsers met RGT criteria and completed PEG IFN-α-2b/RBV treatment at week 24. The remaining three patients had discontinued treatment before the week 24 assessment. Rates of SVR12 and SVR24 for patients stopping treatment at week 24 were 90.9% (20/22) for treatment-naïve patients and 100% (28/28) for prior relapsers.

Virologic response

A rapid decline in mean plasma HCV RNA levels was evident in all patient groups up to week 2 (Fig. 3), by which time most patients had achieved levels below the lower limit of quantification.

Most patients in all three groups achieved RVR (60.0–86.2%; Table 2) and cEVR (79.2–100%; Table 2). All treatment-naïve patients and prior relapsers had undetectable levels of HCV RNA at the end of treatment (Table 2). In prior non-responders, 57.7% of patients had undetectable HCV RNA at end of treatment; all patients in this group had a reduction in HCV RNA from baseline of 1 $\log_{10}$ IU/mL or more at week 4.

Viral breakthrough and viral relapse

No viral breakthrough was observed in treatment-naïve patients or prior relapsers. Six prior non-responders (23.1%) had viral breakthrough (Table 2). Two of these six patients experienced breakthrough at week 8 during the simeprevir treatment period. One patient had viral breakthrough at week 8 after discontinuing simeprevir at week 5 upon meeting virologic stopping criteria. The remaining three patients experienced viral breakthrough during PEG IFN-α-2b/RBV-only treatment (weeks 12–24).

Two treatment-naïve patients experienced viral relapse at week 4 of follow-up. One prior relapser experienced viral relapse at week 24 of follow-up. Four of 15 prior non-responders with undetectable HCV RNA at

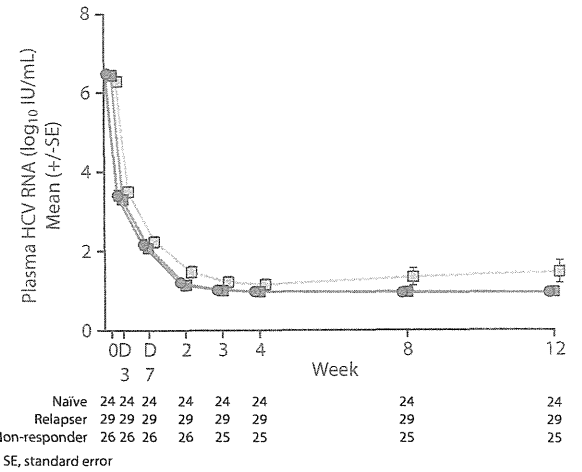

Figure 3 Mean (±SE) change in hepatitis C virus (HCV) RNA levels from baseline to week 12. D, day. —●—, naïve; —■—, relapser; —▪—, non-responder.

Table 2 Virologic response rates

Response, *n*/N (%)	Treatment-naïve ($n = 24$)	Prior relapsers ($n = 29$)	Prior non-responders ($n = 26$)
RVR,† *n* (%)	19/24 (79.2)	25/29 (86.2)	15/25 (60.0)
cEVR,‡ *n* (%)	23/23 (100.0)	28/28 (100.0)	19/24 (79.2)
End of treatment response,§ *n* (%)	24/24 (100.0)	29/29 (100.0)	15/26 (57.7)
Viral breakthrough,¶ *n* (%)	0/24 (0)	0/29 (0)	6/26 (23.1)
Viral relapse,†† *n* (%)	2/24 (8.3)	1/29 (3.4)	4/15 (26.7)

Note: RVR and cEVR are assessed while on treatment. If a subject discontinues all study medications prior to the time point of the parameter of interest, then the subject is not included in the denominator.
†Undetectable HCV RNA at week 4 (i.e. while on treatment). ‡Undetectable HCV RNA at week 12. §Undetectable HCV RNA at end of treatment. ¶An increase of >1.0 $\log_{10}$ IU/mL in HCV RNA level from the lowest level reached, or HCV RNA level of >2.0 $\log_{10}$ IU/mL in patients whose HCV RNA had previously been <1.2 $\log_{10}$ IU/mL detectable or undetectable. ††Detectable HCV RNA during the post-treatment follow-up period of sustained viral response assessment in patients who had undetectable plasma HCV RNA at end of treatment. The incidence of viral relapse was calculated only for patients with undetectable HCV RNA levels at end of treatment and with ≥1 follow-up HCV RNA measurement. The denominator for prior non-responders was $n = 15$.
cEVR, complete early virologic response; HCV, hepatitis C virus; N, number of patients with data at specific time point assessed; *n*, number of patients with observation; RVR, rapid virologic response.

end of treatment had documented viral relapse at the week 4 follow-up visit.

Emerging mutations in treatment failures

Sequencing analysis of the NS3 protease domain of HCV was available for 17 (two treatment-naïve patients, one prior relapser and 14 non-responders) of the 18 simeprevir-treated patients classified as treatment failures (i.e. met predefined virologic stopping criteria, detectable HCV RNA at end of treatment, viral breakthrough or viral relapse). Emerging mutations were identified for 16 patients at the time of failure, including six patients with viral breakthrough. D168V was the most frequent emerging mutation, accounting for eight single mutations, followed by Q80R+D168E (three patients), D168E (two patients), and R155K, D168T and Q80K+D168E (one patient each).

SVR according to selected demographic and baseline disease characteristics

A summary of SVR12 rate by selected demographic and baseline disease characteristics is presented in Table 3. Most treatment-naïve patients and all prior relapsers achieved SVR12; therefore, no apparent trend was noted

Table 3 SVR12 rates by selected demographic and baseline disease characteristics

Characteristic	SVR12 rate, *n*/N (%)		
	Treatment-naïve ($n = 24$)	Prior relapsers ($n = 29$)	Prior non-responders ($n = 26$)
Sex			
Male	7/8 (87.5)	16/16 (100.0)	5/13 (38.5)
Female	15/16 (93.8)	13/13 (100.0)	5/13 (38.5)
Age			
<65 years	19/19 (100.0)	20/20 (100.0)	7/22 (31.8)
≥65 years	3/5 (60.0)	9/9 (100.0)	3/4 (75.0)
IL28B genotype (rs8099917)			
TT	16/16 (100.0)	26/26 (100.0)	0/2 (0.0)
TG/GG	6/8 (75.0)	3/3 (100.0)	10/24 (41.7)
IL28B genotype (rs12979860)			
CC	16/16 (100.0)	26/26 (100.0)	0/2 (0.0)
CT/TT	6/8 (75.0)	3/3 (100.0)	10/24 (41.7)

SVR12, undetectable hepatitis C virus RNA 12 weeks after the last treatment.

in SVR12 rates for the selected demographic and baseline disease characteristics. For non-responders, the number of patients in each subgroup was too small to draw firm conclusions. One prior non-responder who was infected with HCV genotype 1a did not achieve SVR12.

Normalization of ALT

At the end of treatment, the proportion of patients for whom ALT levels were abnormal at baseline and changed to be within the normal limits (based on WHO toxicity grades) was 13/15 (86.7%) for treatment-naïve patients, 8/13 (61.5%) for prior relapsers and 8/13 (61.5%) for prior non-responders.

Safety

Adverse events and laboratory investigations reported as AE during the entire treatment period are summarized in Table 4. All patients experienced at least one AE. No deaths occurred. Two serious AE occurred during treatment – peripheral T-cell lymphoma (unspecified) and hyperbilirubinemia – of which hyperbilirubinemia was considered by the investigator likely to be related to simeprevir. The majority of AE were grade 1 or 2. Grade 3 AE mainly occurred by week 4 and were reported for 17 patients; the most frequent grade 3 AE were neutropenia (6.3%), decreased white blood cell count (5.1%), leukopenia (3.8%) and decreased neutrophil count (3.8%). One patient experienced grade 4 decreased neutrophil count at week 1, which was considered unrelated to simeprevir. No neutropenia-related AE were serious or led to permanent discontinuation of study treatment.

Three patients discontinued treatment due to AE. One patient discontinued all three study medications at week 8 owing to grade 1 anemia, which was considered very likely related to RBV. Two patients discontinued PEG IFN-α-2b/RBV after the simeprevir treatment period owing to grade 3 allergic dermatitis and grade 2 depression, considered probably related to RBV and PEG IFN-α-2b, respectively. Rates of PEG IFN-α-2b and RBV dose interruptions due to AE were 12.7% and 17.7%, respectively, with anemia being the most frequently reported AE leading to dose interruption (5.1% for PEG IFN-α-2b and 7.6% for RBV).

The most common AE reported in more than 30% of patients overall during the entire treatment period were pyrexia (84.8%), anemia (50.6%), decreased white blood cell count (58.2%), malaise (48.1%) and headache (45.6%). During the simeprevir treatment period, the most common AE were pyrexia (83.5%), decreased white blood cell count (58.2%) and malaise (48.1%). There were no differences between patient groups in the incidence of each AE (Table 4).

Rash (any type) was reported in 34 patients (43.0%) during the entire treatment period (25 patients [31.6%] during the simeprevir treatment period), which included rash ($n = 30$; 38.0%), erythema ($n = 6$; 7.6%), skin exfoliation ($n = 2$; 2.5%), erythema multiforme ($n = 1$; 1.3%) and photosensitivity reaction ($n = 1$; 1.3%). All rash AE were of grade 1 or 2 severity. None of these AE were serious or led to permanent discontinuation of the study treatment.

Median values over time for selected laboratory parameters are presented in Fig. 4. Median bilirubin values increased transiently during the first two weeks of simeprevir treatment in all patient groups, but returned to baseline levels by week 16 (i.e. within 4 weeks of the end of the simeprevir treatment period). Median levels between weeks 2 and 16 were slightly higher in prior relapsers. Elevation of bilirubin levels was not associated with increases in ALT or AST. Seven patients experienced grade 3 elevations (>2.5 mg/dL) in blood bilirubin and one patient experienced a grade 4 elevation (>5.0 mg/dL). None of the increased bilirubin-related AE led to permanent discontinuation of study treatment. No changes were noted for uric acid or creatinine.

The incidences of grade 3 or 4 treatment-emergent or worsened laboratory abnormalities were low (occurring in <5% of patients), with the exceptions of decreases in absolute neutrophil count (grade 3, 21.5%; grade 4, 2.5%) and increases in bilirubin (grade 3, 8.9%; grade 4, 1.3%).

DISCUSSION

THIS STUDY INVESTIGATED the efficacy and safety of simeprevir in combination with PEG IFN-α-2b/RBV in a mixed population of both treatment-naïve and treatment-experienced patients chronically infected with HCV genotype 1. The dose of simeprevir (100 mg Q.D. as part of triple therapy) and treatment duration (12 weeks) was chosen based on the results of a phase II, dose and duration ranging study in Japanese treatment-naïve patients infected with genotype 1 HCV (DRAGON study).[29]

In this study, simeprevir 100 mg Q.D. for 12 weeks in combination with PEG IFN-α-2b/RBV (administered for a total of 24 or 48 weeks) demonstrated high rates of SVR12 (91.7–100%) and SVR24 (91.7–96.6%) in treatment-naïve patients and prior relapsers. Although

Table 4 Summary of AE during the entire treatment period and during treatment with simeprevir plus PEG IFN-α-2b/RBV (safety population†)

No. of patients (%)	Entire treatment period				Simeprevir + PEG IFN-α-2b/RBV treatment period			
	Treatment-naïve (n = 24)	Prior relapsers (n = 29)	Prior non-responders (n = 26)	Total (n = 79)	Treatment-naïve (n = 24)	Prior relapsers (n = 29)	Prior non-responders (n = 26)	Total (n = 79)
Discontinuation of any study medication due to AE‡	1 (4.2)	1 (3.4)	1 (3.8)	3 (3.8)	0	1 (3.4)	0	1 (1.3)
Discontinuation of simeprevir alone due to AE	0	0	0	0	0	0	0	0
Temporary interruption of PEG IFN-α-2b due to AE	3 (12.5)	4 (13.8)	3 (11.5)	10 (12.7)	N/A	N/A	N/A	N/A
Dose reduction of PEG IFN-α-2b due to AE	11 (45.8)	4 (13.8)	9 (34.6)	24 (30.4)	N/A	N/A	N/A	N/A
Temporary interruption of RBV due to AE	4 (16.7)	6 (20.7)	4 (15.4)	14 (17.7)	N/A	N/A	N/A	N/A
Dose reduction of RBV due to AE	10 (41.7)	11 (37.9)	9 (34.6)	30 (38.0)	N/A	N/A	N/A	N/A
Any serious AE	1 (4.2)	0	1 (3.8)	2 (2.5)	1 (4.2)	0	0	1 (1.3)
Grade 3/4 AE	6 (25.0)	5 (17.2)	7 (26.9)	18 (22.8)	5 (20.8)	4 (13.8)	5 (19.2)	14 (17.7)
Death	0	0	0	0	0	0	0	0
Common AE§								
Pyrexia	18 (75.0)	27 (93.1)	22 (84.6)	67 (84.8)	17 (70.8)	27 (93.1)	22 (84.6)	66 (83.5)
Decreased white blood cell count	17 (70.8)	16 (55.2)	13 (50.0)	46 (58.2)	17 (70.8)	16 (55.2)	13 (50.0)	46 (58.2)
Anemia	11 (45.8)	21 (72.4)	8 (30.8)	40 (50.6)	11 (45.8)	16 (55.2)	6 (23.1)	33 (41.8)
Malaise	12 (50.0)	12 (41.4)	14 (53.8)	38 (48.1)	12 (50.0)	12 (41.4)	14 (53.8)	38 (48.1)
Headache	11 (45.8)	12 (41.4)	13 (50.0)	36 (45.6)	13 (41.7)	10 (34.5)	12 (46.2)	32 (40.5)
Decreased appetite	12 (50.0)	12 (41.4)	7 (26.9)	31 (39.2)	12 (50.0)	12 (41.4)	7 (26.9)	31 (39.2)
Injection-site reactions	11 (45.8)	8 (27.6)	12 (46.2)	31 (39.2)	10 (41.7)	7 (24.1)	11 (42.3)	28 (35.4)
Rash	12 (50.0)	8 (27.6)	10 (38.5)	30 (38.0)	10 (41.7)	7 (24.1)	8 (30.8)	25 (31.6)
Alopecia	14 (58.3)	9 (31.0)	5 (19.2)	28 (35.4)	Overall incidence <30%			
Arthralgia	11 (45.8)	10 (34.5)	6 (23.1)	27 (34.2)	10 (41.7)	10 (34.5)	6 (23.1)	26 (32.9)
Decreased neutrophil count	11 (45.8)	8 (27.6)	7 (26.9)	26 (32.9)	11 (45.8)	8 (27.6)	7 (26.9)	26 (32.9)
Decreased platelet count	11 (45.8)	6 (20.7)	8 (30.8)	25 (31.6)	11 (45.8)	6 (20.7)	7 (26.9)	24 (30.4)

†All patients who received study drugs. ‡Permanent discontinuation of all study medication (i.e. discontinuation of simeprevir and PEG IFN-α-2b/RBV at the same time, or discontinuation of PEG IFN-α-2b/RBV after completion or discontinuation of simeprevir). §Occurring in >30% patients overall.
AE, adverse events; N/A, not available; PEG IFN, peginterferon; RBV, ribavirin.

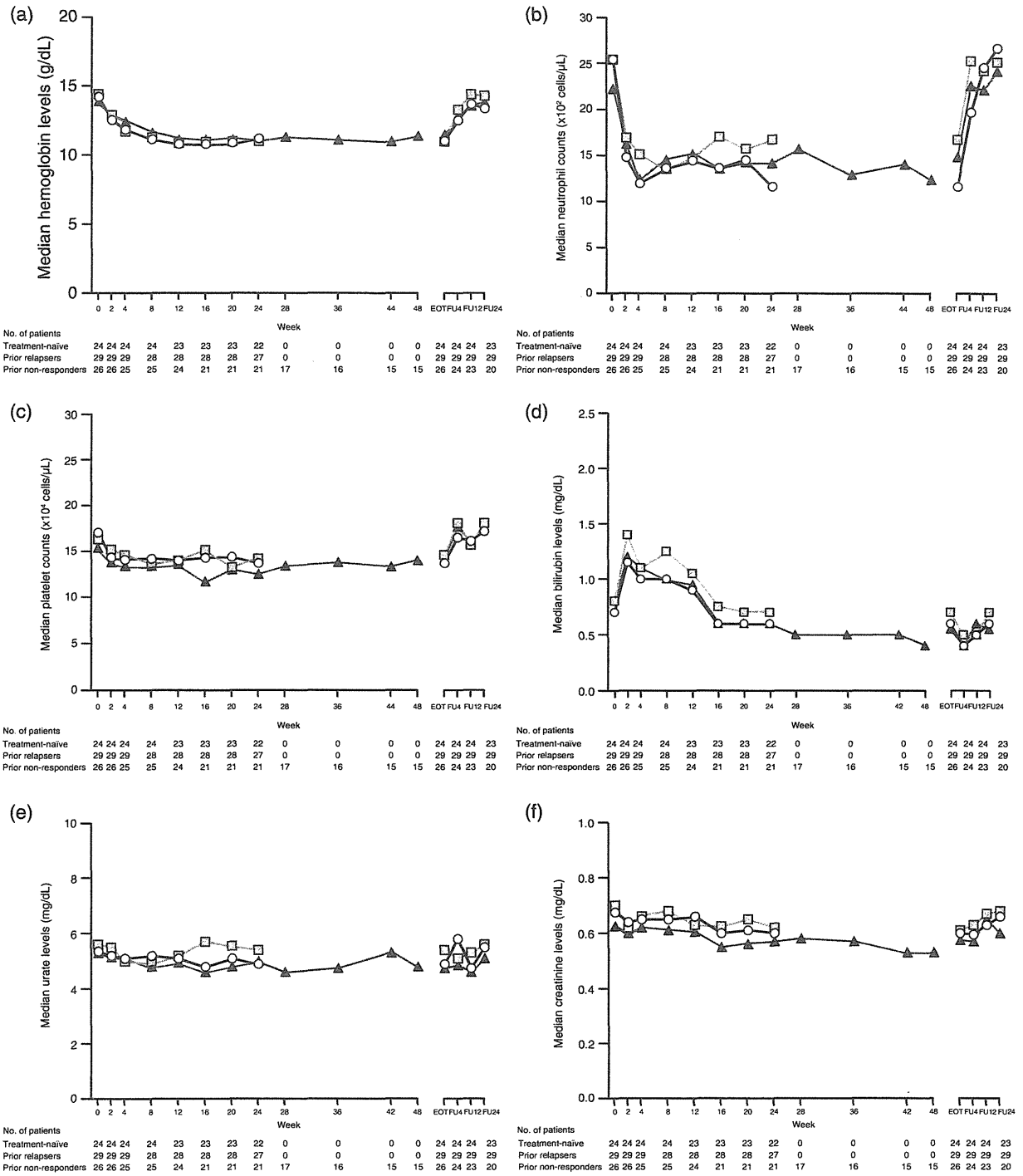

Figure 4 Changes in laboratory parameters over time. (a) Hemoglobin; (b) neutrophil count; (c) platelet count; (d) bilirubin; (e) urate; and (f) creatinine. EOT, end of treatment; FU, follow-up. —○—, treatment-naïve ($n = 24$); □, prior relapsers ($n = 29$); —▲—, prior non-responders ($n = 26$).

our study is somewhat limited by the small patient numbers and non-comparative design, the results in this study population are consistent with the high rates of virologic response that have been reported previously for simeprevir in combination with PEG IFN-α-2a/RBV in other phase III studies in Japan[25,26] and in international studies.[22–24] In QUEST-2, a phase III study conducted outside Japan, simeprevir combined with PEG IFN-α-2a/RBV or PEG IFN-α-2b/RBV was found superior to each PEG IFN-α/RBV regimen alone in treatment-naïve HCV genotype 1 patients.[30] The response rates also compare favorably with those demonstrated in trials of telaprevir in combination with PEG IFN-α-2b/RBV in treatment-naïve patients and prior relapsers.[11,13]

The rates of SVR among prior non-responders who received 48 weeks of treatment with PEG IFN-α-2b/RBV were lower than those for treatment-naïve patients and prior relapsers, consistent with other studies of prior non-responders.[11,16,22] Most patients had received prior treatment with PEG IFN/RBV. A high proportion of the patients in this group had heterozygous TG genotypes for a polymorphism (rs8099917) in the *IL28B* locus that has been associated with poor response to therapy.[31]

All patient groups showed a rapid reduction in HCV RNA levels within the first two weeks of treatment with simeprevir, and most patients achieved RVR at week 4 (60.0–86.2%) and cEVR at week 12 (79.2–100%). All treatment-naïve patients and prior relapsers had undetectable HCV RNA at the end of treatment, with no viral breakthrough and a very low incidence of viral relapse during follow-up in these two groups. Six (23%) of 26 prior non-responders had viral breakthrough, which was documented during simeprevir treatment for three of these patients. Emerging mutations in the NS3 protease domain were identified for isolates from all six patients at the time of breakthrough and for 16 of the 17 patients classified as treatment failures in the study. All patients except for one prior non-responder were infected with HCV genotype 1b. Some of the mutations identified (D168E/T/V, Q80R/K, R155K) have been previously described in HCV genotype 1b isolates following exposure to simeprevir *in vitro* and in clinical studies[23,25,27] and are thought to confer reduced susceptibility. Data from studies of other PI suggest that poor response to IFN-based therapy increases the likelihood of the emergence of resistant isolates.[6] However, further studies are needed to evaluate emerging mutations in HCV genotype 1 and their clinical impact.

In this study, an RGT approach (based on levels of HCV RNA) was used to determine whether treatment-naïve patients or prior relapsers could reduce the duration of PEG IFN-α-2b/RBV therapy to 24 weeks. More than 90% of these patients were able to stop PEG IFN-α-2b/RBV at 24 weeks rather than continuing to 48 weeks. Those patients who stopped therapy at 24 weeks had high rates of SVR12 and SVR24 (90.9–100%), despite the shorter treatment duration. The reduction in exposure to PEG IFN-α-2b/RBV therapy following initial triple combination therapy with simeprevir could potentially limit the extent and duration of PEG IFN-α-2b/RBV-related AE.

Treatment with simeprevir 100 mg Q.D. in combination with PEG IFN-α-2b/RBV was well tolerated, with mostly grade 1/2 AE. Notably, there was a very low incidence of treatment discontinuation due to AE, and those reported were considered to be related to PEG IFN-α-2b and/or RBV. This is in contrast to studies of first-generation PI, which have reported rates of treatment discontinuation due to AE of 10–20%.[11,12,14] In addition, serious toxicities that have been widely reported with first-generation PI (e.g. anemia, cutaneous reactions, neutropenia)[6,12,13,15,32] were mainly of grade 1 or 2 severity in this study of simeprevir. In phase II studies of simeprevir, mild transient hyperbilirubinemia has been reported.[21] Although patients in this study experienced a transient elevation of blood bilirubin, levels returned to baseline values after simeprevir treatment and the elevation was not associated with increases in ALT or AST levels. Also, there were no treatment discontinuations due to increased bilirubin-related AE. Telaprevir has been associated with increases in uric acid and creatinine.[14] No changes were noted in median uric acid and creatinine values in this study of simeprevir.

In conclusion, treatment with simeprevir 100 mg Q.D. for 12 weeks in combination with PEG IFN-α-2b/RBV (for 24 or 48 weeks) demonstrated potent antiviral activity and high rates of SVR in patients who were treatment-naïve or had previously relapsed after IFN-based therapy, with most patients having a shorter treatment duration. Antiviral activity was also demonstrated in patients who had failed to respond to prior IFN-based therapy. Simeprevir was well tolerated in all patients. The present phase III CONCERTO-4 study demonstrates the efficacy and safety of simeprevir in a relatively small sample of treatment-naïve and previously treated patients with chronic HCV genotype 1 infection, while CONCERTO-1, -2 and -3 provide further data on the efficacy and safety of this regimen in a larger population.

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Effect of prolonged administration of pegylated interferon/ribavirin therapy in genotypes 2a and 2b: Propensity score-matched analysis

Satoe Yokoyama,* Yoshiiku Kawakami,*,† Michio Imamura,* C Nelson Hayes,*,† Hiroshi Kohno,‡ Hirotaka Kohno,‡ Keiji Tsuji,§ Yasuyuki Aisaka,¶ Shinsuke Kira,¶ Keitarou Yamashina,** Michihiro Nonaka,†† Shoichi Takahashi,*,† Takashi Moriya,‡‡ Mikiya Kitamoto,§§ Shiomi Aimitsu,** Toshio Nakanishi,†† Hiroiku Kawakami¶¶ and Kazuaki Chayama*,†

*Department of Gastroenterology and Metabolism, Applied Life Sciences, Institute of Biomedical and Health Sciences and †Liver Research Project Center, Hiroshima University, §Department of Gastroenterology, Hiroshima City Asa Hospital, ¶Department of Gastroenterology, Hiroshima Red Cross Hospital, **Department of Gastroenterology, Hiroshima General Hospital of West Japan Railway Company, ††Department of Gastroenterology, Miyoshi Central Hospital, ‡‡Department of Gastroenterology, Chugoku Rousai Hospital, §§Department of Gastroenterology, Hiroshima Prefectural Hospital, ¶¶Department of Gastroenterology, Kawakami Clinic, Hiroshima, and ‡Department of Gastroenterology, Kure Medical Center, Kure, Japan

Key words
genotype 2a, genotype 2b, hepatitis C, pegylated interferon plus ribavirin therapy, prolonged therapy.

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Correspondence
Dr Yoshiiku Kawakami, Department of Medicine and Molecular Science, Division of Frontier Medical Science, Programs for Biomedical Research, Graduate School of Biomedical Sciences, Hiroshima University, 1-2-3 Kasumi, Minami-ku, Hiroshima 734-8551, Japan. Email: kamy4419@hiroshima-u.ac.jp

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Abstract

Background and Aim: Chronic hepatitis C genotype 2 patients show high susceptibility to pegylated interferon plus ribavirin therapy (PEG/RBV). However, the differences in response to therapy between genotypes 2a and 2b, and the efficacy of prolonged therapy for refractory patients have not been evaluated. We investigated the differences in response to PEG/RBV between each genotype and examined the efficacy of prolonged therapy.

Methods: A total of 343 chronic hepatitis patients infected with hepatitis C virus (HCV) genotype 2 (2a: $n = 195$; 2b: $n = 148$) were enrolled in this study. All patients received PEG/RBV for 24 (24 week group, $n = 242$) or more weeks (prolonged group, $n = 101$). We analyzed the differences in virological response between genotypes 2a and 2b. Clinical and virological factors of patients in the 24-week group and the prolonged treatment group were matched using propensity score analysis, and the efficacy of prolonged therapy established by comparing time of serum HCV disappearance for each genotype.

Results: Virological response tended to be higher for genotype 2a compared with genotype 2b; however, there was no significant difference in sustained virological response rates between genotypes (2a: 78.3%; 2b: 70.2%; $P = 0.19$). After propensity score matching, the adjusted P-value for sustained virological response rate was significantly different for genotype 2b patients with undetectable HCV-RNA between weeks 5 and 8, and for genotype 2a patients with detectable HCV-RNA at week 8.

Conclusion: Prolonged therapy with PEG/RBV may be effective when serum HCV-RNA is detectable at week 4 and week 8 for genotype 2b and 2a patients, respectively.

Introduction

Worldwide, approximately 150 million people are persistently infected with hepatitis C virus (HCV), and about 30% of them develop serious liver disease. Every year, more than 350 000 people die from hepatitis C-related liver diseases such as decompensated cirrhosis and hepatocellular carcinoma.[1,2] In Japan, HCV seroprevalence has been reported in 1.5–2.3% of the population,[3] and about 70% of patients with chronic hepatitis C are infected with HCV genotype 1. For the remaining 30% of patients, 20–25% are infected with genotype 2a and 5–10% with genotype 2b.[4]

Standard therapy for patients with high viral loads of HCV genotype 2 is pegylated interferon (PEG-IFN) plus ribavirin (RBV) combination therapy (PEG/RBV) for 24 weeks. It has been reported that 70–93% of HCV genotype 2-infected patients can achieve sustained virological response (SVR) with this therapy.[5] The reported SVR rate for genotype 2-infected patients in Japan was about 80%. A significant difference in SVR rate between genotypes 2a and 2b has not been previously reported. Because of the high response rate to PEG/RBV therapy, the differences between genotype 2a and 2b have not been examined fully. Thus, some articles have discussed reducing therapy duration,[6,7] but prolongation of therapy may be important in about 20% of refractory patients. In Japan, the standard duration for treatment of HCV genotype 2-infected patients is 24 weeks, but recommendations by physicians have resulted in refractory patients undergoing longer

periods of therapy. In this study, we investigated the differences in virological response to PEG/RBV for genotypes 2a and 2b. Furthermore, we compared patients infected with the same genotype who had undergone different durations of therapy after balancing both groups using propensity score matching[8] and examined the efficacy of prolonged therapy for each genotype.

Patients and methods

Patients. A total of 343 chronic hepatitis patients infected with HCV genotype 2 were treated with PEG/RBV at Hiroshima University Hospital or one of 13 other hospitals or clinics in Hiroshima between August 2002 and September 2010. All patients were infected with high titer (> 10 000 IU/mL) HCV genotype 2a or 2b. HCV subtype was identified by direct sequence analysis. Neither genotype 2c nor subsets of genotype 1 were identified in this study. Exclusion criteria included liver disease from another cause, decompensated cirrhosis, presence of liver cancer, hepatitis B or human immunodeficiency virus infection, renal insufficiency, history of heart disease or cerebral infarction, pregnancy, or currently breastfeeding.

Study design. All patients were treated subcutaneously with PEG-IFN-α-2b (Pegintron; Schering-Plough, Tokyo, Japan) once weekly and orally with 600–1200 mg/day RBV (Rebetol; Schering-Plough). PEG-IFN dose was based on the patient's body weight measured at the start of therapy (60 μg for ≤ 45 kg; 80 μg for > 45 kg to ≤ 60 kg; 100 μg for > 60 kg to ≤ 75 kg, and 120 μg for > 75 kg to ≤ 90 kg). RBV dose was also adjusted according to body weight: 600 mg for ≤ 60 kg, 800 mg for > 60 kg to ≤ 80 kg, and 1000 mg for > 80 kg, based on information supplied by the manufacturer. These are the durations and dosages approved by the Japanese Ministry of Health, Labour and Welfare. The daily dose of RBV was reduced by 200 mg when hemoglobin (Hgb) levels fell below 10 g/dL, or for an acute decrease followed by stabilization of Hgb concentrations at more than 3 g/dL below baseline, or for the onset of clinical symptoms of anemia (e.g. palpitations, dyspnea on effort, and fatigue) associated with a decrease in Hgb of > 2 g/dL from baseline. Once the RBV dose was reduced, it was maintained at that level throughout the remainder of the study when patients complained of anemia-related symptoms of fatigue or pallor. RBV was discontinued when Hgb levels fell below 8.5 g/dL, or where patients manifested more severe anemia including orthostatic hypotension; patients exhibiting the latter were excluded from the analysis. Patients who could not continue therapy because of other side-effects, such as flu-like symptoms, fatigue, nausea, vomiting, diarrhea, dizziness, depression, and hair loss, were also excluded from the study.

Data collection. Background factors included age, sex, body weight, history of antiviral therapy, and duration of therapy. Viral load, white blood cell count, Hgb, platelet count, and levels of serum aspartate aminotransferase, alanine aminotransferase, γ-glutamyltranspeptidase, creatinine, HgbA1c, and total cholesterol were analyzed. Patients were followed up for 24 weeks after the cessation of active therapy. Patients underwent HCV-RNA testing at least once every 4 weeks. At each visit, blood samples were collected for hematology and blood chemistry analysis, undertaken at the local hospital laboratory using standard methodologies. HCV-RNA was centrally assessed by qualitative reverse transcription polymerase chain reaction. SVR was defined as undetectable HCV-RNA at 24 weeks after the completion of therapy. The study was conducted in accordance with the Declaration of Helsinki and was approved by the local Ethics Committees of all participating centers. Written informed consent was obtained from all participating patients.

Data management and statistical analysis. The 24-week therapy group (24-week group) comprised patients who had received PEG/RBV for between 20 and 28 weeks. Patients who had undergone therapy for more than 20 weeks were included in the analysis as it was expected they would have received > 80% of the PEG/RBV of patients who had undergone the full 24 weeks of therapy.[9] Despite a treatment period of 20 weeks, there were some cases where PEG/RBV was less than 80% of a sufficient dose. However, there was no difference in background with comparison groups. The prolonged therapy group (prolonged group) comprised patients who had received more than 29 weeks of PEG/RBV therapy.

To analyze the efficacy of prolonged therapy, background differences were adjusted, excluding therapy duration, for the 24-week group and the prolonged group using propensity score analysis. Age, sex, viral load, and history of antiviral therapy were included to control for the influence of confounding covariates. Hgb was also used as a covariate for comparisons of genotype 2b. Propensity score matching was performed using R version 2.14.0 (University of Tsukuba, Ibaraki, Japan). The propensity score matching method used to produce balanced samples was evaluated and found to have a c statistic of 0.67. The estimated SVR rate was established using the inverse probability weighting method. The SVR rate was presumed by the reciprocal of the propensity score. All covariates were assumed to have a strong association with SVR.[10] Continuous variables were tested for normal distribution with the Kolmogorov–Smirnov test. In the case of dichotomous variables, group differences were examined with the Pearson chi-squared or Fisher's exact test, as appropriate. A *P*-value of 0.05 was considered to indicate statistical significance.

Results

Baseline characteristics. The patients' baseline characteristics are shown in Table 1. The 24-week group included 139 HCV genotype 2a and 103 genotype 2b patients. The prolonged therapy group included 56 genotype 2a and 45 genotype 2b patients. HCV viral loads of genotype 2b-infected patients were significantly higher than those of genotype 2a-infected patients in both groups (Table 1). Genotype 2a-infected patients of the prolonged group were older, had lower platelet counts, and had greater anamneses of IFN therapy than for the 24-week group. With the exception of Hgb levels, no other difference in background was observed for genotype 2b patients in either group. Each patient had been assessed by a doctor and were directed to undergo treatment for either 24-weeks or longer. To date, there are no universal guidelines to determine which patients should undergo prolonged

Table 1 Characteristics of genotype 2a and genotype 2b patients in the 24-week and prolonged therapy groups

	24-week group		Prolonged group					*P*-value****
	Genotype 2a (*n* = 185)	Genotype 2b (*n* = 111)	Genotype 2a (*n* = 56)	Genotype 2b (*n* = 45)	*P*-value*	*P*-value**	*P*-value***	
Age (years)	58 (21–79)	57 (17–82)	60 (32–79)	58 (22–82)	0.74	0.11	0.017	0.46
Sex (male/female)	88/97	57/54	25/31	20/25	0.61	0.86	0.70	0.43
BW (kg)	60 (35.7–86)	57.6 (41–95)	57.3 (40–71.6)	57.6 (41–95)	0.67	0.38	0.83	0.61
BMI (kg/m^2)	23.05 (16.1–35.2)	22.8 (16.6–30.3)	22.07 (15.7–35.2)	22.1 (16.6–30.3)	0.37	0.59	0.58	0.38
Histological stage (F0/F1/F2/F3/F4)	7/79/42/21/6	1/50/20/16/2	2/15/8/7/5	0/13/15/4/5	0.37	0.125	0.06	0.38
Prior IFN therapy (-/mono therapy/PEG + RBV)	135/47/3	87/24/0	26/25/5	24/15/6	0.37	0.60	0.25	0.10
WBC (× 10^3/mL)	5000 (2100–13500)	5100 (2660–10100)	4930 (2700–9800)	4800 (1750–11370)	0.88	0.0037	0.69	0.84
Neut (× 10^3/mL)	2580 (857–9544)	2611 (892–7917)	2548 (1291–5949)	2734 (987–4585)	0.62	0.54	0.60	0.79
Hgb (g/dL)	13.9 (8.3–17.5)	14.0 (11.0–15.3)	13.7 (8.4–16.8)	13.6 (10.8–18.5)	0.77	0.50	0.31	0.047
PLT (× 10^4/mm^3)	17.4 (7.8–32.9)	17.6 (4.0–39.5)	15.4 (6–32.9)	17.8 (6.6–38)	0.81	0.14	0.14	0.60
AST (IU/L)	40 (6.9–279)	36 (11–214)	38 (20–175)	33 (13–229)	0.58	0.40	0.40	0.47
ALT (IU/L)	52.5 (11–354)	43 (11–214)	33.5 (13–240)	32 (11–462)	0.486	0.75	0.76	0.17
γ-GTP (IU/L)	27 (8–709)	30 (9–397)	31 (11–199)	26 (11–631)	0.20	0.79	0.777	0.89
HCV-RNA (log IU/mL)	5.95 (5.0–6.98)	6.38 (5.03–7.3)	5.97 (5.13–7.30)	6.44 (5.45–7.12)	< 0.05	0.0015	0.0015	0.41
T-chol (mg/dL)	174 (70–297)	183.5 (112–279)	182 (115–261)	186 (129–235)	0.10	0.93	0.93	0.67
TG (mg/dL)	97 (39–699)	93 (10–590)	99.5 (12–1207)	112 (53–231)	0.085	0.80	0.74	0.20
IL28B (rs8099917)	47/16/1	31/7/1	16/9/0	14/6/0	0.72	0.19	0.61	0.600
PEG > 80%/80%>	116/29	68/10	34/5	29/4	0.18	0.92	0.31	0.92
RBV > 80%/80%>	92/53	50/28	30/10	26/9	0.92	0.94	0.17	0.29

*Genotype 2a *versus* genotype 2b in 24-week PEG/RBV; **genotype 2a *versus* genotype 2b in prolonged PEG/RBV; ***24-week PEG/RBV *versus* prolonged PEG/RBV in genotype 2a; ****24-week PEG/RBV *versus* prolonged PEG/RBV in genotype 2b.

γ-GTP, γ-glutamyl transpeptidase; ALT, alanine aminotransferase; AST, serum aspartate aminotransferase; BMI, body mass index; BW, body weight; HCV, hepatitis C virus; Hgb, hemoglobin; IFN, interferon; Neut, neutrophil cell; PEG/RBV, pegylated interferon plus ribavirin therapy; PLT, platelet count; T-chol, total cholesterol; TG, triglyceride; WBC, white blood cell.

TT: IL28B SNP rs8099917 genotype TT, which is relatively sensitive to IFN therapy. IL28B TG/GG: IL28B SNP rs8099917 genotypes TG/GG, which are relatively resistant to IFN therapy.

Underlined values were significantly different factors between each groups.

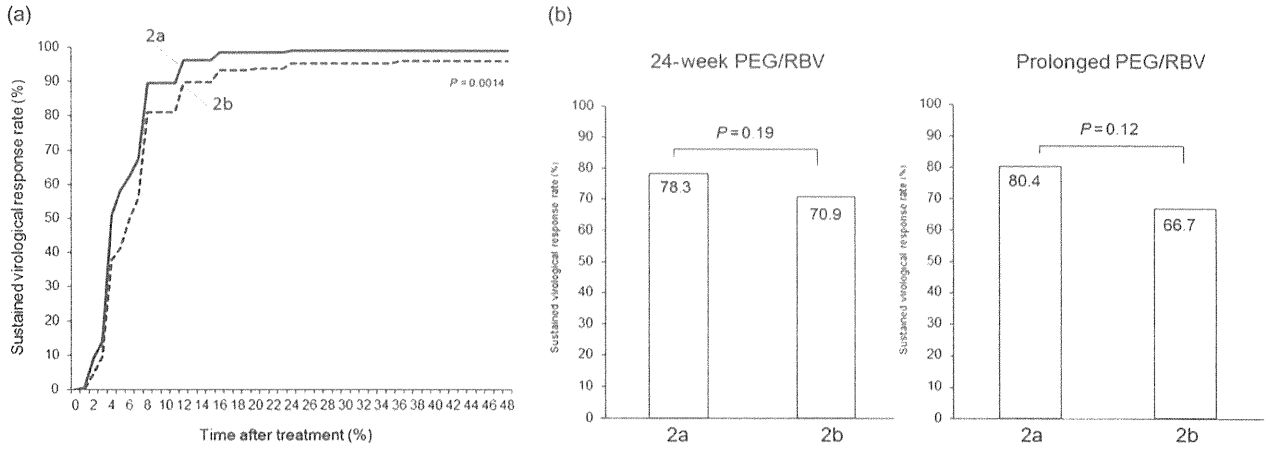

Figure 1 Virological response to pegylated interferon (PEG-IFN) and ribavirin (RBV) combination therapy in hepatitis C virus (HCV) genotype 2a- and 2b-infected patients. (a) Cumulative rate of serum HCV-RNA disappearance. (b) The ratio of sustained virological response for the 24-week and prolonged group. A significant difference between genotype 2a and 2b patients was recognized for the 24-week therapy group ($P = 0.007$ by log-rank analysis). The cumulative rate of HCV-RNA disappearance from weeks 4 to 11 was significantly higher for genotype 2a patients than for genotype 2b ($P < 0.05$ by chi-squared test). By comparison, no significant difference in disappearance rate was recognized between genotype 2a and 2b patients in the prolonged therapy group.

therapy. Therefore, there was a tendency for advanced fibrosis or IFN therapy refractory cases to be present for genotype 2a of the prolonged group.

Differences in virological responses to PEG/RBV therapy revealed cumulative rates for HCV-RNA disappearance were significantly higher for patients infected with genotype 2a compared with genotype 2b ($P = 0.0014$; log rank test in the 24-week group) (Fig. 1a). However, the SVR ratio for each genotype was similar for both the 24-week and prolonged groups (Fig. 1b).

The relationship between time of HCV-RNA disappearance and SVR rate was investigated (Fig. 2). A delay in HCV-RNA disappearance corresponded with a decrease in SVR rate for genotypes 2a and 2b in the 24-week group. For the prolonged group, patient therapy duration was determined independently by their treating physician and was found to be similar for genotype 2a (median 48 [29–72] weeks) and genotype 2b (median 48 [29–96] weeks). For the prolonged group, genotype 2b-infected patients who had undetectable HCV-RNA at week 9 or later had significantly lower SVR rates compared with patients with undetectable HCV-RNA by 8 weeks. However, for genotype 2a patients undergoing prolonged therapy, the SVR rate dropped gradually and was not considered statistically significant.

Finally, the efficacy of prolonged therapy for each genotype was investigated. Patients who achieved a rapid virological response (RVR) were more likely to achieve a SVR, and therefore a total of 154 patients (2a: 98, 2b: 44) with undetectable HCV-RNA at 4 weeks were excluded from this analysis. The remaining 189 patients (2a: 97, 2b: 92) were divided into two groups based on whether serum HCV-RNA disappeared by week 8 of therapy. We used propensity scores to match the clinical and virological factors of patients in the 24-week group and compared the SVR rate according to time of HCV-RNA disappearance for each genotype. Significant differences in background factors such as sex, IFN anamnesis, and white blood count were abolished after propensity score matching (Table 2). Prolonged therapy coincided with an increase in SVR rate for genotype 2a patients who were HCV-RNA positive at week 8 of therapy and for genotype 2b patients who were HCV-RNA negative at week 8 (Fig. 3).

Discussion

It is well known that the therapeutic effects of PEG/RBV treatment of patients with chronic hepatitis C varies according to HCV genotype.[11] Because of the high SVR rate, therapy guidelines stipulate that genotype 2 patients undergo therapy for 24 weeks.[12] Mangia *et al.* reported that 12 weeks of therapy was sufficient for patients who achieved a RVR and that it was efficacious as standard therapy.[6] For patients without a RVR, the SVR rate following therapy was reported to be 48–77%; hence, additional prolonged therapy is required for refractory patients.[13] In previous reports, no significant difference in SVR rate was observed between 24 (81%) and 48 weeks (76–80%) of PEG/RBV therapy.[14,15] These studies included both HCV genotype 2 and 3 patients. Zeuzem *et al.* reported that following 24 weeks of therapy, SVR rates for genotype 2 (93%) and genotype 3 (79%) patients were not significantly different.[5] There was some association between rs12979860 and liver histopathology for genotype 3 patients but not for genotype 2 patients,[16] so further investigation is warranted. Arase *et al.* believe that prolonged therapy is effective for genotype 1 patients,[17] while others have shown that prolonged therapy does not improve the SVR rate for genotype 2 patients.[18] Currently, prolonged therapy is performed only for patients who have demonstrated a tolerance for continuous therapy. In previous studies, genotypes 2a and 2b were investigated collectively as genotype 2, with few reports investigating them individually. The purpose of this study was to examine whether the therapeutic effects of PEG/RBV were similar for genotype 2a and 2b patients, and whether prolonged therapy was effective for each of these genotypes.

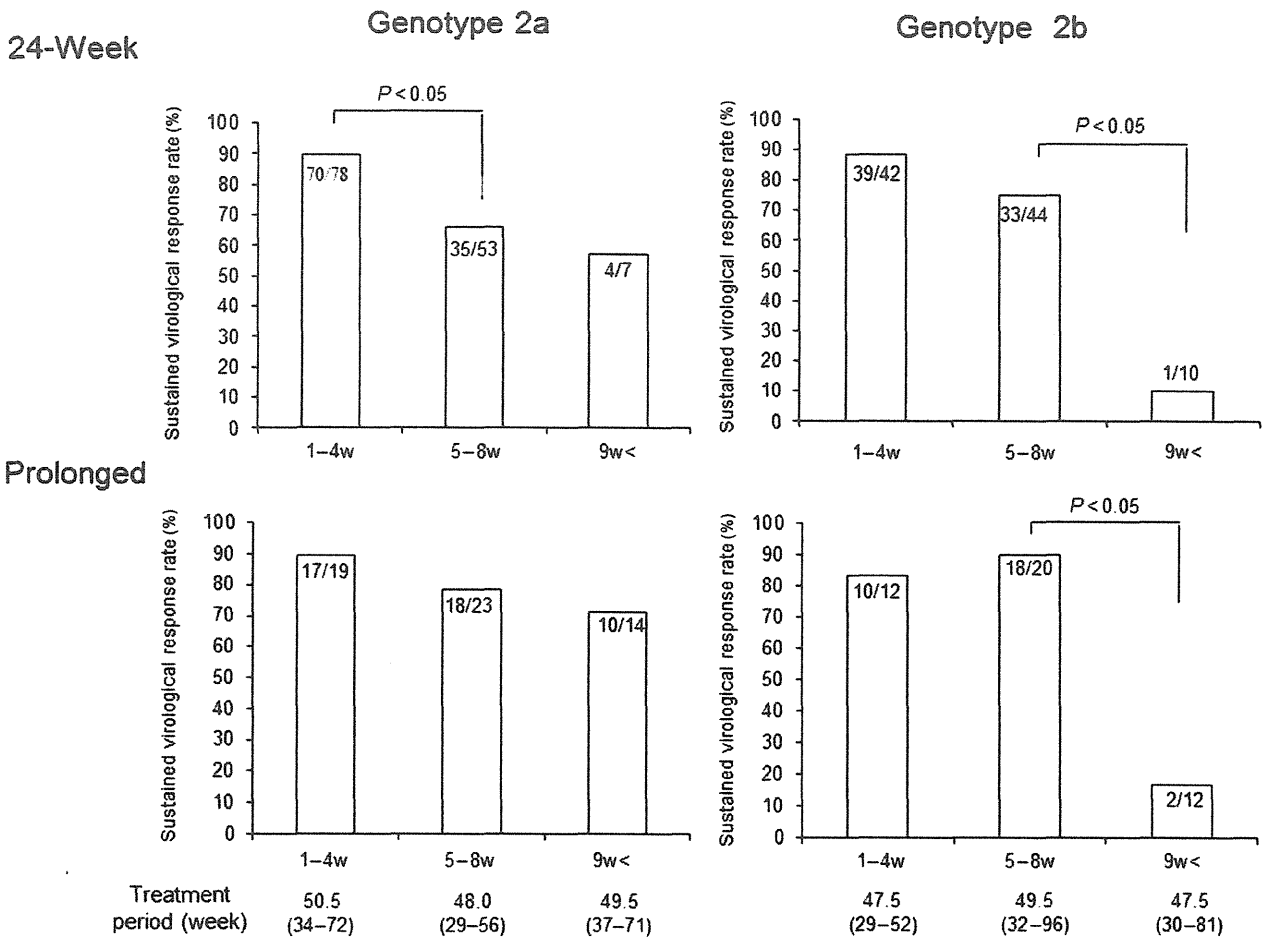

Figure 2 Sustained virological response rate according to the time point of hepatitis C virus (HCV)-RNA disappearance in the 24-week and prolonged therapy group. When HCV-RNA disappearance was delayed, as for the 24-week therapy group, the sustained virological response (SVR) rate gradually decreased. In particular, the SVR rate for genotype 2b patients, for HCV-RNA disappearance at > 9 weeks, was considerably lower than for genotype 2a patients. The SVR rate improved for both genotype 2a and 2b patients when the HCV negative period was prolonged.

In our study, the disappearance rate of HCV-RNA between weeks 4 and 11 was significantly higher for genotype 2a than for genotype 2b (Fig. 1a). Higher SVR rates were reported for genotype 2a-infected patients compared with genotype 2b-infected patients in other studies,[14,19] but no such difference was observed in the current study (Fig. 1b).

Therefore, our results may reflect differences because of other factors, such as viral load, which was lower for genotype 2a compared with genotype 2b patients. SVR rates corresponding to time point serum HCV-RNA disappearance differed between the 24-week and prolonged group for both subtypes (Fig. 2). The SVR rate was more than 80% for RVR patients for both subtypes and for both therapy groups. Non-RVR patients in the prolonged group tended to have higher SVR rates than those in the 24-week group for both subtypes. However, differences in background characteristics of patients may have led to these results. Significant differences in viral load and patient age were observed for genotype 2a patients between the two groups, while Hgb levels were different for genotype 2b patients (Table 2). To control for this bias, we used propensity scores to compare the two groups after excluding RVR patients. Using this method, we found that prolonged therapy was effective for genotype 2a patients who were HCV-RNA positive at week 8 and genotype 2b patients who were HCV-RNA negative by week 8. The average treatment duration for patients in the prolonged group was 48 weeks. Nagoshi *et al.* reported that SVR rates tend to increase when the HCV-RNA negative period is greater than 17 weeks.[20] Protracted therapy duration may improve SVR rates for genotype 2b patients who are HCV-RNA negative for relatively short periods; alternatively, it may be necessary to consider other therapies. Further investigation is required.

Previously, only one report has examined the difference in viral sequence heterogeneity between the two genotypes and their response to IFN. The presence of four or more nucleotide mutations in the IFN/RBV resistance-determining region (IRRDR) of HCV 2a was significantly associated with RVR and SVR. In genotype 2b-infected patients, the presence of two or more nucleotide