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RAPID COMMUNICATION

Daclatasvir Plus Asunaprevir for Chronic HCV Genotype 1b Infection

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All-oral combinations of direct-acting antivirals may improve efficacy and safety outcomes for patients with hepatitis C virus (HCV) infection, particularly those who are poor candidates for current interferon/ribavirin-based regimens. In this open-label, phase 3 study, 135 interferon-ineligible/intolerant and 87 nonresponder patients with chronic HCV genotype 1b infection were enrolled at 24 centers in Japan. Patients received daclatasvir 60 mg once daily plus asunaprevir 100 mg twice daily for 24 weeks. The primary endpoint was sustained virologic response 24 weeks after treatment (SVR_{24}). This study is registered with Clinical Trials.gov (NCT01497834). SVR_{24} was achieved by 87.4% of interferon-ineligible/intolerant patients and 80.5% of nonresponder (null and partial) patients; rates were similar in cirrhosis (90.9%) and noncirrhosis (84.0%) patients, and in patients with *IL28B* CC (84.5%) or non-CC (84.8%) genotypes. Fourteen patients in each group (12.6%) discontinued dual therapy, mainly due to adverse events or lack of efficacy. Nine nonresponder patients received additional treatment with peginterferon/ribavirin per protocol-defined criteria. The rate of serious adverse events was low (5.9%) and varied among patients. The most common adverse events were nasopharyngitis, increased alanine aminotransferase (ALT) and aspartate aminotransferase (AST), headache, diarrhea, and pyrexia. *Conclusion:* Interferon-free, ribavirin-free all-oral therapy with daclatasvir and asunaprevir for 24 weeks is well tolerated and can achieve a high rate of SVR in patients with HCV genotype 1b who were ineligible, intolerant, or had not responded to prior interferon-based therapy. (HEPATOLOGY 2014;59:2083-2091)

Treatment of chronic hepatitis C virus (HCV) infection typically includes a regimen of interferon-based therapy plus ribavirin, with or without a direct-acting antiviral. The efficacy and tolerability of these regimens are not ideal, and there remains a large number of patients for whom these treatments are not acceptable or viable. The addition of direct-acting antivirals can improve treatment outcomes for patients infected with chronic HCV. When combined with peginterferon and ribavirin, the HCV protease inhibitors telaprevir, boceprevir, or simeprevir achieved overall sustained virologic response (SVR) rates ranging from 68% to 89% in treatment-naïve patients with HCV genotype 1 infection.[1-3] Patients with no response to previous peginterferon/ribavirin therapy did not respond as well to this combined regimen, with rates of SVR ranging from 34% to 52%.[3-5] In Japan, patients chronically infected with HCV are older and predominantly infected with HCV genotype 1, both factors which impact response to therapy.[6] For Japanese patients who had no prior response to treatment with peginterferon/ribavirin, telaprevir or

Abbreviations: HCV, hepatitis C virus; LLOQ, lower limit of quantitation; NS, nonstructural; SVR, sustained virologic response; TD, target detected; TND, target not detected.

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simeprevir plus peginterferon and ribavirin provided SVR rates of only 34% or 38-51%, respectively.[3,7] The array of adverse events associated with peginterferon and ribavirin is well known; incremental toxicities associated with the addition of telaprevir to peginterferon and ribavirin included anemia, skin disorders and severe rash, and gastrointestinal-related disorders, while the addition of simeprevir is associated with hyperbilirubinemia due to inhibition of hepatic bilirubin transporters.[3] For patients who cannot tolerate or are not eligible for treatment with interferon-based therapy because of coexisting morbidities, treatment options are few to none. Clearly, the current treatment options are not adequate and an urgent unmet need remains for better treatment regimens for these patient populations.

Daclatasvir is a first-in-class, NS5A replication complex inhibitor with potent pan-genotypic antiviral activity *in vitro* (HCV genotypes 1-6).[8] Asunaprevir is a potent, selective NS3 protease inhibitor with antiviral activity against HCV genotypes 1, 4, 5, and 6 *in vitro*.[9] Both daclatasvir and asunaprevir have demonstrated robust antiviral activity, with no clinically meaningful pharmacokinetic interactions between them when coadministered.[8,10,11] Preliminary phase 2 studies showed potent antiviral effects using daclatasvir and asunaprevir as an all-oral therapy and in combination with a regimen of peginterferon/ribavirin in patients infected with HCV genotype 1 who had not responded to prior therapy.[12,13] We evaluated the safety and antiviral activity of interferon-free, ribavirin-free, all-oral therapy with daclatasvir and asunaprevir in a phase 3 trial involving Japanese patients infected with HCV genotype 1b who are interferon-ineligible/intolerant or nonresponders (null and partial) to interferon-based therapies.

Materials and Methods

Patients. A total of 259 patients were enrolled at 24 centers in Japan from January 5 2012 to March 30 2012. Eligible patients were men and women, 20 to 75 years of age, with chronic HCV genotype 1b infection, an HCV RNA level of 10^5 IU/mL or higher, with a body-mass index of 16 to 35 kg/m^2, and, in up to 10% of enrolled patients, evidence of compensated cirrhosis (Child-Pugh A), as documented either by liver biopsy or discriminated by a previously described algorithm.[14]

Key exclusion criteria included evidence of hepatocellular carcinoma, coinfection with hepatitis B virus or human immunodeficiency virus, or previous exposure to inhibitors of NS5A or NS3 protease. Patients with alanine aminotransferase (ALT) of more than 5 times the upper limit of normal range, total bilirubin of 2 mg/dL or higher, an international normalized ratio of 1.7 or higher, an albumin level 3.5 g/dL or below, and a platelet count of less than 50,000/mm^3 were also excluded.

Patients ineligible for interferon-based therapy, but potentially eligible for enrolment in this study, were treatment-naïve and considered poor candidates for interferon-based therapy because of medical complications including anemia, neutropenia, thrombocytopenia, depression, advanced age ($\geq$65 years), or other conditions deemed not suitable for interferon-based therapy by the investigator, including hypertension, diabetes mellitus, autoimmune disease, and abnormal thyroid function. Patients intolerant to interferon-based therapy had received interferon-based therapy for less than 12 weeks and previously discontinued from therapy due to toxicities associated with interferon or ribavirin. Patients who were null or partial responders to previous peginterferon/ribavirin or interferon-beta/ribavirin therapy were defined as never having attained an undetectable HCV RNA level after at least 12 weeks of therapy. Null responders included patients who never attained at least a 2-$\log_{10}$ decrease from baseline in HCV RNA levels at week 12, and partial responders never achieved undetectable HCV RNA levels after 12 weeks of therapy.

Study Design. In this open-label, phase 3 study of two patient cohorts, interferon-ineligible/intolerant and

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nonresponder patients received daclatasvir and asunaprevir for 24 weeks. Patients were followed for an additional 24 weeks after treatment. Daclatasvir was administered orally at a dose of 60 mg once daily, and asunaprevir was administered orally at a dose of 100 mg twice daily. Host *IL28B* genotype was assayed for the rs12979860 single-nucleotide polymorphism by Monogram Biosciences using a real-time polymerase chain reaction (PCR) assay.

Nonresponder patients who met futility criteria, defined as an increase in viral load of at least 1 $\log_{10}$ or confirmed detectable HCV RNA of at least 15 IU/mL on or after week 8, were eligible for addition of peginterferon-alpha/ribavirin to continued treatment with daclatasvir and asunaprevir for an additional 24 weeks at the discretion of the investigator. Interferon-ineligible/intolerant patients were not candidates for interferon-based therapy; therefore, daclatasvir/asunaprevir dual therapy was stopped if futility criteria were met.

Study Oversight. This study was approved by the Institutional Review Board at each participating site and was conducted in compliance with the Declaration of Helsinki, Good Clinical Practice guidelines, and local regulatory requirements. All patients provided written informed consent.

Efficacy Assessments. HCV RNA levels were measured using the Roche COBAS Taqman test with a lower and an upper limit of quantitation of 15 IU/mL and 6.9 $\times$ 10^7 IU/mL, respectively. HCV RNA was measured at screening and at day 1, weeks 1, 2, 4, 6, 8, 10, 12, 16, 20, and 24, and posttreatment at weeks 4, 8, 12, and 24.

Resistance Testing. Patient-derived HCV NS5A and NS3/4A sequence populations were PCR-amplified and sequenced. Patient samples selected for sequencing included all baseline samples and samples from patients with virologic failure.

Safety Assessments. Safety evaluations included reported adverse events and serious adverse events, clinical laboratory tests, physical examinations, and electrocardiograms.

Endpoints. The primary efficacy endpoint was the proportion of patients with HCV RNA <15 IU/mL (target detected [TD] or target not detected [TND]) at 24 weeks after completion of daclatasvir and asunaprevir treatment, including patients who discontinued treatment early. Key secondary endpoints included the proportion of patients with undetectable HCV RNA (TND) at weeks 4 and 12, at the end of treatment, and HCV RNA <15 IU/mL (TD or TND) at 12 weeks after the end of treatment. Safety endpoints included the frequency of serious adverse events, adverse events, discontinuations due to adverse events, and laboratory abnormalities.

Statistical Analysis. Analyses included all patients who received at least one dose of study medications. For virologic response, 2-sided 95% confidence intervals were calculated based on the normal approximation to the binomial distribution. Categorical variables were summarized using counts and percents. Continuous variables were summarized with univariate statistics. Patients with missing data or those who received additional peginterferon/ribavirin therapy were considered failures.

Role of the Funding Source. The study was designed and conducted by the sponsor (Bristol-Myers Squibb/Bristol-Myers KK) in collaboration with the principal investigators. The sponsor collected the data, monitored the study conduct, and performed the statistical analyses. All authors had access to the data and assume responsibility for the accuracy, integrity, and completeness of the reported data and for the fidelity of this report to the trial protocol. The article was prepared by authors employed by Bristol-Myers Squibb, with input from all authors and the assistance of a medical writer employed by Bristol-Myers Squibb. All authors made the decision to submit the article for publication.

Results

Patients. In all, 222 patients received treatment, 135 in the interferon-ineligible/intolerant group (100 medically ineligible for interferon, 35 intolerant to interferon) and 87 in the nonresponder group (48 null responders, 36 partial responders, 3 undetermined) (Fig. 1). Demographic baseline characteristics of patients are shown in Table 1. As expected, when compared with reported demographics from U.S. and European studies, patients were older and a larger proportion were female. Similar to the global population, however, there were more patients with *IL28B* CC genotype in the interferon-ineligible/intolerant population (69.6%) and more patients with *IL28B* non-CC genotype in the nonresponder population (81.6%). Overall, the rate of discontinuations from dual therapy was low (12.6%; 14 patients in each group), and was due primarily to adverse events (nine patients [6.7%] in the interferon-ineligible/intolerant group, two patients [2.3%] in the nonresponder group) and lack of efficacy (four patients [3.0%] in the interferon-ineligible/intolerant group, 11 patients [12.6%] in the nonresponder group).

Virologic Response. HCV RNA levels declined rapidly after initiation of treatment in both groups (Fig. 2). At week 2, the mean decrease in HCV RNA

Table 1. Demographic and Baseline Characteristics of Patients and Their Disease

Characteristic	Interferon-Ineligible/Intolerant n = 135	Nonresponder n = 87	Total N = 222
Age, years			
- Median	64.0	60.0	62.5
- Range	24-75	42-74	24-75
- ≥65 years, n (%)	62 (45.9)	27 (31.0)	89 (40.1)
Male sex, n (%)	38 (28.1)	39 (44.8)	77 (34.7)
IL28B rs12979860 genotype, n (%)			
- CC	94 (69.6)	16 (18.4)	110 (49.5)
- CT	40 (29.6)	66 (75.9)	106 (47.7)
- TT	1 (0.7)	5 (5.7)	6 (2.7)
HCV RNA			
- Mean $\log_{10}$ IU/mL ± SD	6.6 ± 0.58	6.8 ± 0.47	6.6 ± 0.55
- ≥800,000 IU/mL, n (%)	109 (80.7)	80 (92.0)	189 (85.1)
Cirrhosis, n (%)	11 (8.1)	11 (12.6)	22 (9.9)
Response to prior therapy (nonresponders), n (%)			
- Null	NA	48 (55.2)	48 (21.6)
- Partial	NA	36 (41.4)	36 (16.2)
- Other	NA	3 (3.4)*	3 (1.4)
Premedical status (interferon-ineligible/intolerant), n (%)			
- Ineligible-naïve	100 (74.1)	NA	100 (45.0)
• Depression	10 (10.0)	NA	10 (10.0)
• Anemia/neutropenia/thrombocytopenia	44 (44.0)	NA	44 (44.0)
• Other complications requiring medications†	34 (34.0)	NA	34 (34.0)
• Advanced age	12 (12.0)	NA	12 (12.0)
- Intolerant	35 (25.9)	NA	35 (15.8)

*Three patients had insufficient data to be classified as partial or null nonresponders.
†Other complications included hypertension, diabetes mellitus, autoimmune disease, abnormal thyroid function, insomnia, stroke, and psychological.
NA = not applicable.

from baseline was 5.2 $\log_{10}$ IU/mL. Overall, 167/222 patients (75.2%) had undetectable HCV RNA at week 4 during treatment, and 202 patients (91.0%) had undetectable HCV RNA at week 12 on treatment. At 12 weeks after the end of treatment period, 119 (88.1%) interferon-ineligible/intolerant and 70 (80.5%) nonresponder patients had achieved SVR_{12}; by 24 weeks after the end of treatment 118 (87.4%) interferon-ineligible/intolerant and 70 (80.5%) nonresponder patients had achieved SVR_{24} (Table 2). Patients with cirrhosis also achieved high rates of SVR_{24} (20/22, 90.9%). When analyzed by *IL28B* genotype, the response rates were similar for patients with *IL28B* CC genotype (84.5%) and *IL28B* non-CC genotypes (84.8%) (Table 2). Other baseline factors including gender, age, and baseline HCV RNA, did not appear to impact response rates (Table 2).

Virologic Failure. Thirty-four (15.3%) patients (17 each in the interferon-ineligible/intolerant group and nonresponder group) were considered virologic failures. Of patients with undetectable HCV RNA at the end of treatment, 11/129 (8.5%) interferon-ineligible/intolerant patients experienced viral relapse during posttreatment follow-up. Six of 76 patients (7.9%) in the nonresponder group with undetectable HCV RNA at the end of treatment had viral relapse. Two patients in the interferon-ineligible/intolerant group and one patient in the nonresponder group had detectable HCV RNA at the end of treatment. Virologic breakthrough occurred in 4 (3.0%) interferon-ineligible/intolerant patients and in 10 (11.5%) nonresponder patients. At the discretion of the investigators, 9 of the 10 nonresponder patients with virologic breakthrough had additional treatment with peginterferon/ribavirin according to protocol-defined criteria; all nine patients were declared treatment failures in the analysis of the primary endpoint. One of the nine patients who received additional peginterferon/ribavirin responded to treatment with no detectable HCV RNA at follow-up week 24, two patients had HCV RNA detectable at end of treatment, and six patients relapsed.

Of the 34 patients with virologic failure, 29 had resistance-associated substitutions to both daclatasvir (predominantly NS5A-L31M/V-Y93H) and asunaprevir (predominantly NS3-D168 variants) detected at failure. Twenty-two patients with virologic failure had NS5A polymorphisms L31M/V and/or Y93H prior to treatment (Supporting Table 1).

We also investigated the influence of pretreatment resistance-associated variants on efficacy in this study. Pretreatment L31M, Y93H, or linked L31V+Y93H NS5A polymorphisms were detected in 7, 29, and 1 of the 214 patients with available baseline NS5A

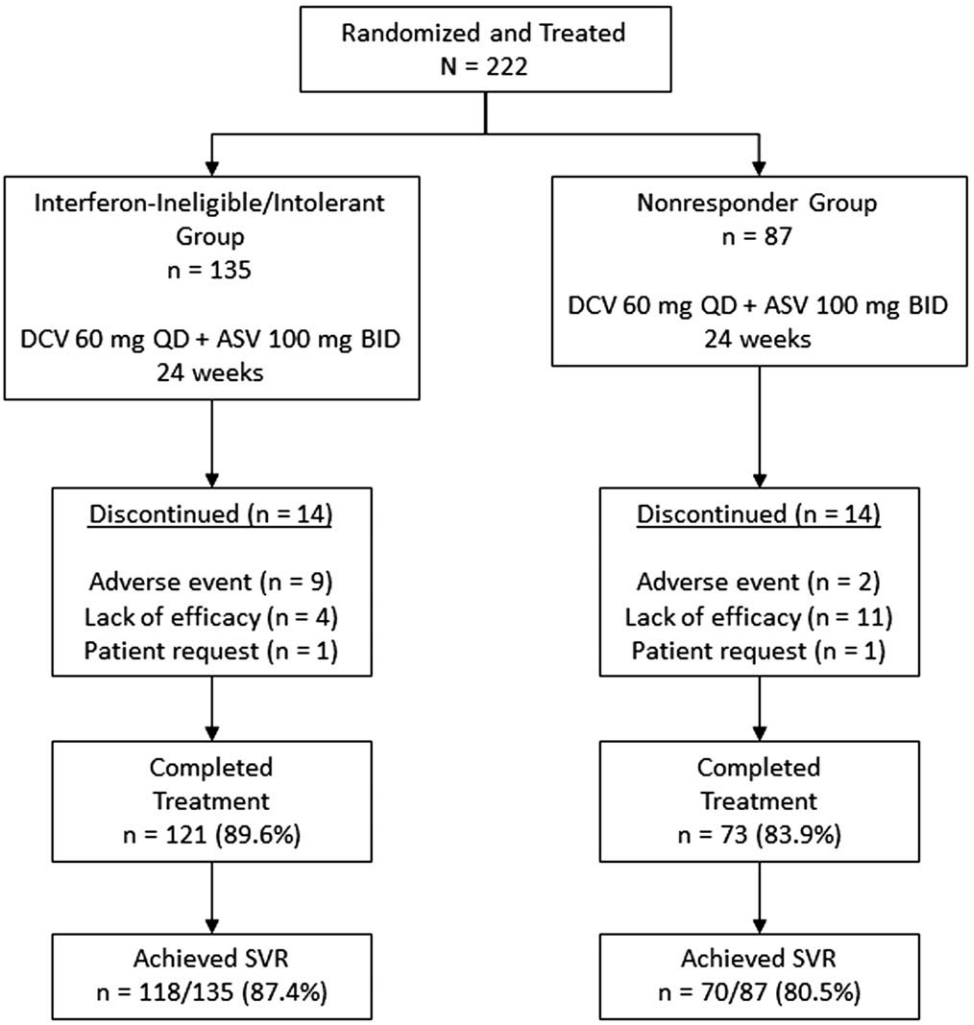

Fig. 1. Patient disposition.

sequences, respectively. Of the 37 patients with L31M/V and/or Y93H at baseline, 11/23 interferon-ineligible/intolerant patients and 4/14 nonresponder patients achieved SVR. The primary asunaprevir resistance-associated variant, NS3-D168E, was present in 2/221 patients with available baseline NS3 sequences; neither of these patients had concomitant NS5A resistance-associated variants. One of these patients achieved SVR; the other relapsed posttreatment.

In comparison with patients who achieved SVR, patients with virologic failure were more likely to have daclatasvir and asunaprevir trough concentrations below their respective median values but within the expected range (Supporting Fig. 1). Most patients with trough concentrations below median values achieved SVR. Treatment compliance, assessed by pill counts and interviews at each study visit, was 83.9% in prior nonresponders and 88.9% in interferon-ineligible/intolerant patients. Across both cohorts, patients with $\geq$95% compliance in dose and duration of treatment had an SVR_{24} rate of 92.7% (179/193), compared with a 31.0% (9/29) SVR_{24} rate in patients who were <95% compliant (15 out of the 29 patients were discontinued due to the lack of efficacy).

Safety. A total of 194 patients (87.4%) completed 24 weeks of therapy, 121 (89.6%) in the interferon-ineligible/intolerant group and 73 (83.9%) in the nonresponder group. No deaths occurred during the study period. Eleven patients (5.0%) discontinued after 4 to 23 weeks of treatment; 10 discontinued due to ALT and aspartate aminotransferase (AST) elevations and one patient discontinued due to myasthenia gravis, with subsequent detection of preexisting myasthenia gravis-related antibodies.

The most common adverse events were nasopharyngitis, increased ALT and AST, headache, diarrhea, and pyrexia (Table 3). Serious adverse events were reported in 13 (5.9%) patients during treatment. In nine (6.7%) interferon-ineligible/intolerant patients, these events included periarthritis, schizoaffective disorder, myasthenia gravis, myocardial infarction, pyrexia, appendicitis, pyelonephritis,

Table 2. Virologic Outcomes

Virologic Response, n (%) [95% CI]	Interferon-Ineligible/Intolerant n = 135	Nonresponder n = 87	Total N = 222
Week 4,*	114 (84.4)	53 (60.9)	167 (75.2)
	[78.3, 90.6]	[50.7, 71.2]	[69.5, 80.9]
Week 12,*	125 (92.6)	77 (88.5)	202 (91.0)
	[88.2, 97.0]	[81.8, 95.2]	[87.2, 94.8]
Weeks 4 and 12,*	106 (78.5)	48 (55.2)	154 (69.4)
	[71.6, 85.4]	[44.7, 65.6]	[63.3, 75.4]
End of treatment response*	129 (95.6)	76 (87.4)	205 (92.3)
	[92.1, 99.0]	[80.4, 94.3]	[88.8, 95.8]
Sustained virologic response	126 (93.3)	71 (81.6)	197 (88.7)
4 weeks after treatment (SVR_4)†	[89.1, 97.5]	[73.5, 89.7]	[84.6, 92.9]
Sustained virologic response	119 (88.1)	70 (80.5)	189 (85.1)
12 weeks after treatment (SVR_{12})†	[82.7, 93.6]	[72.1, 88.8]	[80.5, 89.8]
Sustained virologic response	118 (87.4)	70 (80.5)	188 (84.7)
24 weeks after treatment (SVR_{24})†	[81.8, 93.0]	[72.1, 88.8]	[79.9, 89.4]
SVR_{24} by subpopulations			
- Null responders	N/A	39/48 (81.3)	39/48 (81.3)
- Partial responders	N/A	28/36 (77.8)	28/36 (77.8)
- Undetermined	N/A	3/3 (100)	3/3 (100)
- Ineligible-naïve	85/100 (85.0)	N/A	85/100 (85.0)
- Intolerant	33/35 (94.3)	N/A	33/35 (94.3)
- Cirrhosis	10/11 (90.9)	10/11 (90.9)	20/22 (90.9)
- Noncirrhosis	108/124 (87.1)	60/76 (78.9)	168/200 (84.0)
- Male	32/38 (84.2)	32/39 (82.1)	64/77 (83.1)
- Female	86/97 (88.7)	38/48 (79.2)	124/145 (85.5)
- Age $<$ 65 years	61/73 (83.6)	47/60 (78.3)	108/133 (81.2)
- Age $\geq$ 65 years	57/62 (91.9)	23/27 (85.2)	80/89 (89.9)
- HCV RNA $<$ 800,000 IU/mL	25/26 (96.2)	6/7 (85.7)	31/33 (93.9)
- HCV RNA $\geq$ 800,000 IU/mL	93/109 (85.3)	64/80 (80.0)	157/189 (83.1)
SVR_{24} by *IL28B* genotype (rs12979860)			
- CC	79/94 (84.0)	14/16 (87.5)	93/110 (84.5)
- CT	38/40 (95.0)	52/66 (78.8)	90/106 (84.9)
- TT	1/1 (100)	4/5 (80)	5/6 (83.3)
Virologic failures			
- Virologic breakthrough	4 (3.0)	10 (11.5)‡	14 (6.3)
- End of treatment detectable	2 (1.5)	1 (1.1)	3 (1.4)
- Relapse (among patients undetectable at end of treatment)	11/129 (8.5)	6/76 (7.9)	17/205 (8.3)

*HCV RNA $<$LLOQ ($<$15 IU/mL), target not detected.

†HCV RNA $<$LLOQ, target detected or target not detected.

‡9/10 patients received additional treatment with peginterferon/ribavirin according to protocol criteria.

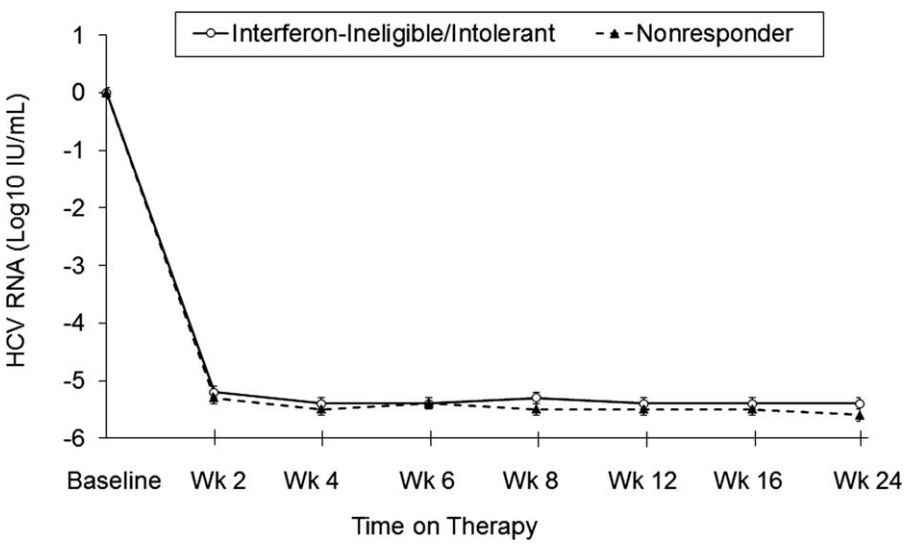

Fig. 2. Mean change in HCV RNA during treatment with daclatasvir and asunaprevir in interferon-ineligible/intolerant and nonresponder patients.

Table 3. Adverse Events and Grade 3-4 Laboratory Abnormalities During the Treatment Period

Event or Laboratory Abnormality, n (%)	Interferon-Ineligible/Intolerant n = 135	Nonresponder n = 87	Total N = 222
Serious adverse events (on treatment)	9 (6.7)	4 (4.6)	13 (5.9)
Adverse event*			
Nasopharyngitis	40 (29.6)	27 (31.0)	67 (30.2)
Increased ALT	24 (17.8)	11 (12.6)	35 (15.8)
Increased AST	18 (13.3)	10 (11.5)	28 (12.6)
Headache	18 (13.3)	17 (19.5)	35 (15.8)
Diarrhea	12 (8.9)	10 (11.5)	22 (9.9)
Pyrexia	12 (8.9)	15 (17.2)	27 (12.2)
Grade 3-4 laboratory abnormality			
Alanine aminotransferase	12 (8.9)	4 (4.6)	16 (7.2)
Aspartate aminotransferase	10 (7.4)	2 (2.3)	12 (5.4)
Hemoglobin	6 (4.4)	1 (1.1)	7 (3.2)
Lymphocytes	5 (3.7)	1 (1.1)	6 (2.7)
Platelets	2 (1.5)	2 (2.3)	4 (1.8)
Bilirubin, total	1 (0.7)	1 (1.1)	2 (0.9)
Neutrophils	0	1 (1.1)	1 (0.5)
Creatinine	1 (0.7)	0	1 (0.5)
Lipase, total	1 (0.7)	0	1 (0.5)

*Adverse events that occurred in more than 10% of patients in any group.

basal cell carcinoma, and hepatocellular carcinoma, respectively; events in four (4.6%) nonresponder patients included second-degree burn, increased liver enzymes, esophageal variceal hemorrhage, and herpes zoster.

ALT and AST elevations were the most frequent adverse events and grade 3/4 laboratory abnormalities (Table 3) and were the basis for 10 of the 11 discontinuations due to adverse events. Two of these 10 patients also had grade 3/4 total bilirubin elevations, but no patient experienced hepatic decompensation. Eight of the 10 patients who discontinued due to ALT/AST elevations (80%) subsequently achieved SVR. For the 16 patients who had grade 3/4 ALT elevations on-treatment, the median time to elevation was ~10 weeks (range 4 to 23 weeks), with rapid reversal in ~2.5 weeks after discontinuation. Most patients with baseline ALT and AST elevations experienced rapid improvement during the first 2 to 4 weeks of treatment, including all patients with grade 3/4 elevations at baseline, with mean decreases at 4 weeks of 43.7 U/L and 35.1 U/L, respectively.

Discussion

Treatment with interferon-based therapy is not an option for many patients with chronic HCV. The findings from this phase 3 study evaluating interferon-free, ribavirin-free, all-oral treatment with daclatasvir and asunaprevir demonstrated high rates of SVR in Japanese patients infected with HCV genotype 1b. Both interferon-ineligible/intolerant and previously treated nonresponder patient groups experienced a rapid reduction in HCV RNA by week 2. The primary endpoint, SVR_{24}, was achieved in 87.4% of patients who were ineligible or intolerant to interferon-based therapies and in 80.5% of patients who had not responded to treatment previously. These high rates of SVR obtained with daclatasvir and asunaprevir represent a significant improvement of cure rates in patient populations typically associated with poor responses to other therapies or with limited therapeutic options. Other factors typically associated with a poor response to therapy, including male gender, high baseline HCV RNA, advanced age, non-CC *IL28B* genotype, and cirrhosis, did not appear to impact response rates, although the number of patients in these subgroups was small.

The response rates in this study were higher than those observed in a phase 3 study evaluating triple therapy with telaprevir and peginterferon/ribavirin in Japanese patients infected with HCV genotype 1 with no response to prior treatment. The SVR rate of nonresponder patients in that study was 34.4%, and safety issues included anemia, severe rash, renal toxicity, and gastrointestinal-related disorders.[7] In a global phase 3 trial, SVR rates ranged from 54% to 59% in partial-responder and 29% to 33% in null-responder patients receiving telaprevir combined with peginterferon/ribavirin.[4] Simeprevir in combination with peginterferon/ribavirin achieved an SVR rate of 38-51% in Japanese nonresponder patients. In the present study, partial-responder and null-responder patients achieved better outcomes (77.8% and 81.3%, respectively), with a much more favorable safety profile. The response rate observed in the ineligible/intolerant group in this study was also notable, especially when considering these patients had no option for curative treatment.

This study was limited to Japanese patients; an ongoing phase 3 study in a similar patient population in the U.S. and Europe will determine whether region-related differences in patient characteristics influence outcomes with this regimen. The results from this phase 3 trial are consistent with the results of a small phase 2a study of Japanese patients treated with daclatasvir and asunaprevir; SVR rates were 64% in peginterferon/ribavirin-ineligible or intolerant patients and 91% in null responder patients.[15] A phase 2b trial combining NS3 (faldaprevir) and NS5B (deleobuvir) inhibitors showed only a 57% SVR in previously untreated patients with HCV genotype-1b infection.[16] In addition, other all-oral regimens earlier in clinical development may provide greater efficacy: in a phase 2

study, SVR rates of 95% to 100% were achieved in treatment-naïve and experienced genotype 1-infected patients treated with sofosbuvir (NS5B inhibitor) in combination with ledipasvir (NS5A inhibitor), with or without ribavirin.[17] The more complex combination of NS3 (ABT-450, plus ritonavir to improve drug exposure), NS5A (ABT-267), and NS5B (ABT-333) inhibitors, with or without ribavirin, achieved SVR rates of 88-96% in treatment-naïve patients and prior null responders with genotype 1 infection. Recent press reports indicate similar results in phase 3 studies with both of these regimens, although full study details are not yet available.[19,20] The combination of daclatasvir and sofosbuvir achieved SVR rates of 88-100% in treatment-naïve patients with genotype 1, 2, or 3 infection, and 95-100% in treatment-experienced patients with genotype 1 infection.[21] However, none of these studies involved patient populations directly comparable to those reported in the present study. Previous experience with HCV regimens indicates that both treatment eligibility and outcomes can vary in relation to variables such as disease stage, patient ethnicity, concomitant medical conditions, and other factors.[3] Further studies of all-oral combinations may provide the evidence needed for optimizing regimen selection on the basis of virologic and patient characteristics.

Response rates at on-treatment week 4 were somewhat higher in the ineligible/intolerant group than in prior nonresponders (84.4% versus 60.9%), but this difference diminished as treatment continued. The early difference in response rates may reflect a reduced contribution of endogenous interferon response in prior nonresponders; the ultimate achievement of an 80.5% SVR rate in this group suggests that such nonresponsiveness can be largely overcome with a potent antiviral regimen.

All-oral treatment with daclatasvir and asunaprevir generally suppressed the enrichment/selection of NS5A and NS3 resistance-associated variants. Virologic failure occurred in 17 patients in each group. Both NS5A and NS3 resistance-associated variants were detected in most patients with virologic failure. There was no apparent association between preexisting NS3 resistance-associated polymorphisms and subsequent virologic outcome. Although more patients with NS5A L31M/V and/or Y93H resistance-associated variants experienced virologic failure, 15/37 of patients with these baseline variants achieved SVR. Thus, pretreatment resistance-associated variants were not absolutely predictive of virologic outcome. Moreover, factors other than resistance, such as lower drug exposure and suboptimal compliance to treatment, likely contributed to treatment failure. The patients with daclatasvir and asunaprevir trough plasma concentrations below median values appeared to be at increased risk of virologic failure (Supporting Fig. 1). Given that patients with $\geq$95% compliance had an SVR_{24} rate of 92.7%, the maintenance of higher compliance is essential for optimizing treatment outcomes.

The rate of premature discontinuation of treatment with daclatasvir and asunaprevir due to adverse events was low. Despite early discontinuation that occurred between weeks 4 and 23, 8 of the 10 patients who discontinued because of elevated levels of ALT and AST achieved SVR_{24}, with rapid reversal of transaminase elevations posttreatment. Although small in number, six patients who achieved SVR were on treatment for 12 weeks or less, suggesting that a shorter treatment period may be possible in some patients. Additionally, baseline elevations of ALT and AST corrected rapidly in most patients after 2 to 4 weeks on treatment, as would be expected with the rapid reduction in HCV RNA levels. The rate of serious adverse events was low and varied among patients, with no consistent pattern of events. The frequency of adverse events was also low, especially compared with historical data in patients receiving a triple regimen with telaprevir and peginterferon/ribavirin that showed a high rate of anemia (91%), pyrexia (85%), and skin disorders (82%).[7]

In conclusion, our findings suggest that 24-week treatment with daclatasvir and asunaprevir provides a highly effective option for patients who currently have no effective treatment options (ineligible or intolerant to interferon-based therapy) and for those patients who did not achieve SVR with prior treatment.

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Contributors: HK, EH, HI, AD, and HM designed the study; EH was the medical lead. HK, YS, KI, JT, YKar, KC, YKaw, AI, KY, KT, NI, KK, TT, NK, and MK recruited patients and obtained the data. HM, HI, EH, and AD analyzed the data. TE and FM provided pharmacokinetic and resistance analyses, respectively. HK, YS, KI, JT, YKar, KC, YKaw, AI, KY, KT, NI, KK, TT, NK, MS, HM, TE, FM, AD,

HI, and EH interpreted study findings. All authors participated in writing the report.

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