

Exposure-Response (Efficacy) Analysis of Daclatasvir and Asunaprevir in Japanese Patients With Hepatitis C Virus Infection

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Abstract

The treatment of hepatitis C virus (HCV) infection has been revolutionized by the development of all-oral combination regimens of direct-acting antiviral agents. The current analysis characterized the relationship between exposures of daclatasvir (DCV; tablets) and asunaprevir (ASV; capsules) and sustained virologic response (SVR) in Japanese patients who are HCV genotype (GT) 1b nonresponders to pegylated interferon (IFN) α /ribavirin or IFN β /ribavirin, and IFN-based therapy–ineligible naive/intolerant patients receiving DCV and ASV, and provided insight into patient covariates that were most closely associated with efficacy. The relationship between the probability of achieving SVR at 12 weeks after treatment (SVR₁₂) and average steady-state plasma concentrations estimated from population pharmacokinetic models for DCV and ASV is described using a logistic regression model with data from a phase 2 and a phase 3 study in Japanese patients infected with HCV GT 1b (N=265). The functional form characterization, which describes a relationship between DCV and ASV average steady-state plasma concentrations and SVR₁₂, as well as covariate identification (demographic, laboratory, and prognostic and treatment covariates) were investigated during model development. The presence of the signature nonstructural protein 5A Y93H mutation at baseline was the only significant parameter of SVR₁₂ in the final exposure-response model. Model evaluation plots demonstrate that the final model was able to predict the observed SVR rates. Exposure-response analysis supports the clinical utility of the combination regimen of 60-mg once-daily DCV and 100-mg twice-daily ASV in Japanese patients infected with HCV GT 1b.

Keywords

hepatitis C virus, direct-acting antivirals, asunaprevir, daclatasvir, exposure-response

In the last 5 years, treatment of chronic hepatitis C virus (HCV) infection has been revolutionized by the development of all-oral combination regimens of direct-acting antivirals (DAAs) that have rapidly superseded pegylated interferons (pegIFNs) plus ribavirin (RBV) as the standard of care.¹ These DAA regimens have proven to be better tolerated than pegIFN-based therapies, with higher rates of posttreatment sustained virologic response (SVR; a surrogate of cure) and shorter treatment durations.^{2,3}

Daclatasvir (DCV; tablets) is an orally available small molecule and pangenotypic nonstructural protein (NS) 5A inhibitor with picomolar in vitro activity against HCV genotypes (GT) 1–6⁴; asunaprevir (ASV; capsules) is also an orally available small molecule and tripeptidic acylsulfonamide inhibitor of the HCV NS3/4A protease with in vitro antiviral activity against GTs 1, 4, 5, and 6.⁵ In early replicon studies, an additive to synergistic interaction between DCV and ASV was observed. DCV and ASV are 2 such DAAs with extensive clinical data in GT 1 infection. The combination therapy of DCV and ASV (DUAL) was approved in Japan in July 2014 as the first all-oral combination therapy for the treatment of patients infected with HCV GT 1; it has also been launched in Korea and other countries.

The combination of DCV once-daily plus ASV twice-daily for 24 weeks has demonstrated high SVR rates in patients infected with the GT 1b subtype of GT 1 in a number of clinical studies, both globally and in East Asian countries where GT 1b infection is predominant.^{6–12} A bioavailability study to select the phase 3 formulation (AI447024) was conducted and, based on these data, a lipid-based softgel capsule was selected. The data from the bioavailability study indicated that the area under the plasma drug concentration-time curve (AUC) of the softgel capsule either with a meal or fasted was approximately twice that of the phase 2 tablet administered with a meal.¹³ The NS5A amino acid polymorphisms L31M/V and

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Y93H have been identified as the major pretreatment HCV resistance-associated variants (RAVs) to DCV in GT 1b, and have been observed to reduce the efficacy of DUAL treatment when present at baseline.¹¹

The objectives of the current paper were to characterize the relationship between the exposures of DCV and ASV predicted by population pharmacokinetic (popPK) models and SVR at posttreatment week 12 (SVR12) as a measure of efficacy in HCV GT 1b-infected Japanese patients who are nonresponders to pegIFN α /RBV or IFN β /RBV and IFN-based therapy–ineligible naive/intolerant and to provide insight into patient covariates that were most closely associated with efficacy based on data from the phase 2 (AI447017)¹¹ and phase 3 (AI447026)⁸ studies.

Method

This study was conducted in accordance with ethical principles that have their origin in the Declaration of Helsinki. The study protocols were approved by all institutional review boards prior to initiation of studies and a written informed consent was obtained from each patient prior to study participation.

Patients and Studies

This exposure-response (E-R) analysis was performed with data from 2 studies; phase 2 study (AI447017) and phase 3 study (AI447026). Study AI447017 was an open-label, phase 2 study in Japanese patients with GT 1b chronic HCV infection to assess safety and tolerability of the regimen of DCV 60 mg once daily plus ASV 600 mg twice daily or 200 mg twice daily using a tablet formulation and to determine the proportion of patients without cirrhosis who were prior null responders to pegIFN α /RBV therapy or ineligible-naive/intolerant to IFN/RBV who achieved SVR, as determined by the investigator. All patients received DUAL therapy for 24 weeks. Study AI447026 was an open-label, phase 3 study to evaluate and assess antiviral activity in nonresponders (null and partial responders) to pegIFN α /RBV or IFN β /RBV and IFN-based therapy–ineligible naive/intolerant patients who were infected with HCV GT 1b. All patients received 60 mg of DCV once daily (tablet formulation) and 100 mg of ASV twice daily (softgel capsule formulation) in combination for 24 weeks and followed for 24 weeks, regardless of HCV RNA status at the end of treatment. The 100-mg softgel capsule, with or without food, was expected to produce similar AUC as that of the 200-mg tablet formulation with food.¹³ SVR12 was a binary variable that indicated HCV RNA below quantifiable limit (BLQ), target detected or target not detected at follow-up week 12 (after end of treatment). HCV RNA was measured using the Roche COBAS TaqMan

(Pleasanton, California) HCV autoassay at all visits. The lower and upper limit of quantification of the assay were 15 IU/mL and 6.9×10^7 IU/mL, respectively.

Exposure-response end point and model development

The efficacy E-R model was developed to describe the relationship between exposures of ASV and DCV using average steady-state plasma concentrations ($C_{av,ss}$) and SVR12. The binary efficacy end point of SVR12, defined as HCV RNA below the limit of detection, used modified intent-to-treat data. The SVR12 rate was assessed on all patients who received at least 1 dose of DUAL treatment (treated subjects). Subjects with missing posttreatment week 12 data were considered nonresponders for computation of the SVR12 rate. However, a subject with a missing posttreatment week 12 HCV RNA measurement was imputed as a responder (SVR12) if the HCV RNA measurements at the scheduled prior visit (posttreatment week 8) and subsequent visit (posttreatment week 24) were BLQ, target detected or not detected. DCV and ASV exposure were assessed as geometric mean of $C_{av,ss}$ and predicted from previously described popPK models. The E-R analysis dataset was prepared by merging $C_{av,ss}$ predicted from the final popPK model for ASV and DCV with SAS (SAS Institute, Inc., Cary, North Carolina) datasets derived from clinical data. These popPK models were developed with the data from phase 2 and phase 3 studies in Japanese subjects with HCV infection and adequately describe the PK profiles of DCV and ASV. The magnitude of estimated covariate effects on DCV PK were small and not clinically meaningful. ASV apparent total body clearance of the drug from plasma decreased with cirrhosis and increasing baseline and time-varying aspartate aminotransferase, indicating an association between hepatic markers and ASV apparent total body clearance of the drug from plasma.¹⁴ (Mayu Osawa, manuscript in preparation/under review).

The covariates of interest investigated within the model were age, body weight, sex, baseline alanine aminotransferase (ALT) level, baseline creatinine clearance (CrCL), *IL28B* GT (rs12979860), NS5A RAV Y93H, baseline HCV RNA, prior treatment status (null or partial responder, or IFN-based therapy ineligible naive/intolerant), cirrhosis status, study (AI447017 or AI447026) and organic anion transporter polypeptide 1B1 (OATP1B1) haplotype.

The E-R relationship was described using a logistic regression model. The probability that a patient achieved SVR12 ($P[SVR12]$) was characterized using a binary logistic regression, such as

$$P(SVR12) = \frac{e^{\mu}}{1 + e^{\mu}}$$

where μ is the logit transform of $P(\text{SVR12})$. The logit (log-odds) is given by

$$\mu = \log \frac{P}{1 - P} = \beta_0 + \beta_i X_i$$

where β is a parameter vector representing the effect of the predictor variable vector X_i on the logit of achieving SVR12, where X_i consists of the covariate (predictor) values for each patient. The functional form relating predictor variable (X_i) and logit of the binary response (μ) were tested as linear, log-linear, or nonlinear relationships.

Model development was conducted in 3 stages. First, the base model was developed to establish the relationship (with an appropriate functional form) between ASV and DCV exposures and SVR12, without consideration of any potential effects of covariates. Given the observed high SVR12 rates across the range of ASV and DCV exposures^{7,9} in the analysis dataset, DCV and ASV $C_{av,ss}$ and the term describing their interactions were prespecified to be included in the base model regardless of their statistical significance to allow for identification of potential covariate exposure interactions. Secondly, the full model was developed to quantify covariate effect. OATP1B1 haplotypes were tested in a univariate fashion on the base model before testing the full model. ASV is a substrate of OATP1B1 and the effect of OATP1B1 haplotypes on ASV exposure was assessed in the popPK analysis. As the results of popPK analysis, the OATP1B1 haplotypes did not show statistical significance, suggesting that in the Japanese population, genetic variability of OATP1B1 has no impact on ASV exposure. In the E-R analysis, OATP1B1 haplotypes were also tested to check if they had an effect on efficacy in the first step of the development of a full model. Because they did not affect ASV exposure, no confounding was expected. However, to avoid over-parameterization, OATP haplotypes were assessed for significance as covariate as the first step, then a full model was developed by incorporating all the other covariates into the base model. A final model was obtained by retaining the statistically significant predictors that potentially modulated the E-R, described by the full model using a backward elimination method. Models were assessed by the likelihood ratio test (LRT) for nested models ($P < .01$, corresponds to objective function value increases of 6.63, 9.21, and 11.34 for 1, 2, and 3 degrees of freedom, respectively), and by the Bayesian information criteria (BIC)¹⁵ for non-nested models. 95% confidence intervals (CIs) of estimated parameters were calculated by bootstrap method. Overview of E-R model development is shown in Supplementary Figure 1.

Exposure-response model evaluations

Model evaluation was conducted using visual predictive check (VPC) based on the final model and was presented stratified by significant covariates. 1000 trials were simulated with observed data based on final model estimated SVR12 rates. The 95% prediction intervals of the SVR12 response rates were summarized from these 1000 trials by each bin, categorized by quartiles of $C_{av,ss}$ and compared with the observed proportion of each bin. In addition, the model-predicted probability (95%CI obtained by bootstrap) of SVR12 was presented with model prediction intervals and observed proportion. The final model was evaluated by assessing the agreement between the observed proportion of SVR12 and the 95% model prediction intervals.

Analysis platforms

Data assembly and modifications were performed using SAS (version 9.2) and final datasets were generated as an SAS transport file. Model development was performed using NONMEM (version 7.2, GloboMax; Hanover, Maryland).

Diagnostic graphics, exploratory analysis, and post-processing of NONMEM output were performed using SAS and S-plus (version 9.2 for SAS, version 8.2 for S-plus).

Results

Patient Characteristics

A total of 265 patients were enrolled in the studies. The patient demographics and characteristics assessed as covariates are presented in Table 1. The median age was 62 years (range, 24-75), median baseline body weight was 55 kg (range, 36-93), 66% of patients were female, and 59% of patients were IFN-ineligible naive or intolerant. Baseline NS5A-Y93H RAV was observed in 15% of patients. The median (range) of $C_{av,ss}$ was 557 (148.8-1486.0) ng/mL for DCV and 164 (59.3-947.1) ng/mL for ASV. Observed SVR12 rates were generally comparable across strata for age, sex, patient group, cirrhosis status, and baseline HCV RNA group, although rates were significantly lower in patients with baseline NS5A-Y93H (47.5%) RAV than in patients without the signature RAV (90.3%; Figure 1). Observed DCV and ASV exposures in patients who did or did not achieve SVR12 were generally comparable (Figure 2). Similarly, DCV and ASV exposures overlapped considerably across baseline RAV and SVR12 status; however, numerical differences were observed (Figure 3).

Model Development

First, the base model was assessed with respect to linear function of ASV and DCV exposures separately or together and with intercept. The effect of interaction

Table 1. Summary of Baseline Demographics and Characteristics Assessed as Covariates (N = 265)

Covariate	Value
Age, median years (range)	62 (24-75)
Weight, median kg (range)	55.0 (36.0-93.4)
Sex	
Male, n (%)	91 (34.3)
Female, n (%)	174 (65.7)
Patient group	
Nonresponder, n (%)	108 (40.8)
IFN ineligible naive/intolerant, n (%)	157 (59.2)
Study	
Phase 2 (AI447017), n (%)	43 (16.2)
Phase 3 (AI447026), n (%)	222 (83.8)
Cirrhosis no/yes, n (%)	243 (91.7)/22 (8.3)
Baseline viral load log ₁₀ , median IU/mL (range)	6.8 (4.9-7.7)
Baseline ALT, median U/L (range)	55.0 (13-377)
<i>IL28B</i> genotype	
CC/TT/CT	129 (48.68)/130 (49.06)/6 (2.26)
NS5A Y93H resistance mutation no/yes/missing, n (%)	217 (81.9)/40 (15.1)/8 (3.0)
Baseline creatinine clearance (mL/min)	84.6 (39.5-172.8)
DCV + ASV $C_{av,ss}$, median ng/mL (range)	
DCV (60 mg once daily)	557.4 (148.8-1486.0)
ASV (tablet 600 mg twice daily)	590.2 (379.4-2111.2)
ASV (tablet 200 mg twice daily)	268.4 (91.0-440.3)
ASV (softgel 100 mg twice daily)	163.7 (59.3-947.1)
OATP haplotype	
*1B/*1B, n (%)	51 (19.2)
*1B/*1A, n (%)	86 (32.5)
*1A/*1A, n (%)	28 (10.6)
Other, n (%)	64 (24.1)
Missing, n (%)	36 (13.6)

ALT, alanine aminotransferase; IL, interleukin; OATP, organic anion transporting polypeptide.

between ASV and DCV was also assessed. In this step, a model including the interaction of both compounds provided a better description of the data. Second, in function assessment, a linear model for both agents had a lower BIC value compared with log-linear and nonlinear function of $C_{av,ss}$, and the results indicated that the model that accounts for linear function, with interaction between ASV and DCV and without intercept, had the lowest BIC. This model provided evidence of a relationship between exposures and P(SVR12) and suggests that a linear model provided the better description. In the model with intercept, the relative standard error of intercept was high (63%) and the intercept was not significant in model development. In addition, the interaction between patient type and exposure were not significant.

Therefore, a linear logistic regression model with $C_{av,ss}$ for both ASV and DCV and interaction between exposures of both compounds as the predictor variable was selected as the final base model, as it provided an adequate and best parsimonious fit to the data

for SVR12. The base model parameter estimates for SVR12 are presented in Supplementary Table 2.

To develop the full model, first the OATP1B1 haplotype was tested and found not statistically significant at a 1% level of LRT relative to the base model and was therefore not retained in the model. These results indicate that genetic variability based on OATP1B1 polymorphisms had no influence on efficacy. Subsequently, the full model was developed by incorporating all covariates as listed in Table 1 into the base model. The full model included the following covariates: age, body weight, sex, baseline ALT, baseline CrCL, *IL28B* GT, Y93H baseline resistance, baseline viral load (log₁₀ IU/mL), patient type, cirrhosis, and study. The continuous covariates were tested with normalization to median for each covariate.

The full model for SVR12 was able to achieve successful minimization and convergence. The full model parameter estimates of all the variables tested for SVR12 are summarized in Supplementary Table 3.

Logit expression for the full model was given by:

$$\begin{aligned} \mu = & \beta_1 \times \text{Cavgss, ASV} + \beta_2 \times \text{Cavgss, DCV} + \beta_3 \\ & \times \text{Cavgss, ASV} \times \text{Cavgss, DCV} + \text{CON}_1 \\ & \times \frac{(\text{BVLV} - 6.8)}{6.8} + \text{CON}_2 \times \frac{(\text{AGE} - 62)}{62} + \text{CON}_3 \\ & \times \frac{(\text{ALT} - 55)}{55} + \text{CON}_4 \times \frac{(\text{WT} - 55)}{55} + \text{CON}_5 \\ & \times \frac{(\text{CRCL} - 84.6)}{84.6} + \text{CAT}_1 \times \text{PATG} + \text{CAT}_2 \\ & \times \text{Y93H1} + \text{CAT}_3 \times \text{Y93H2} + \text{CAT}_4 \\ & \times \text{IL28B1} + \text{CAT}_5 \times \text{GEN} + \text{CAT}_6 \\ & \times \text{SF} + \text{CAT}_7 \times \text{CIRR} \end{aligned}$$

where BVLV is baseline viral load, PATG is patient type, Y93H1 is patients with the Y93H mutation, Y93H2 is patients for whom the Y93H resistance data are missing, *IL28B1* is *IL28B* GT (rs12979860), GEN is sex, SF is study, CIRR is cirrhosis status. CON and CAT represent the effects of each covariate. The variables are median of the respective continuous covariates.

Covariate relationships were considered to be statistically significant provided that the relationship was significant at a 1% level of LRT relative to the full model. Based on this criterion, only the Y93H baseline mutation was retained in the final model. For the other covariates, the prespecified statistical criteria were not sufficient to be retained. The final model parameter

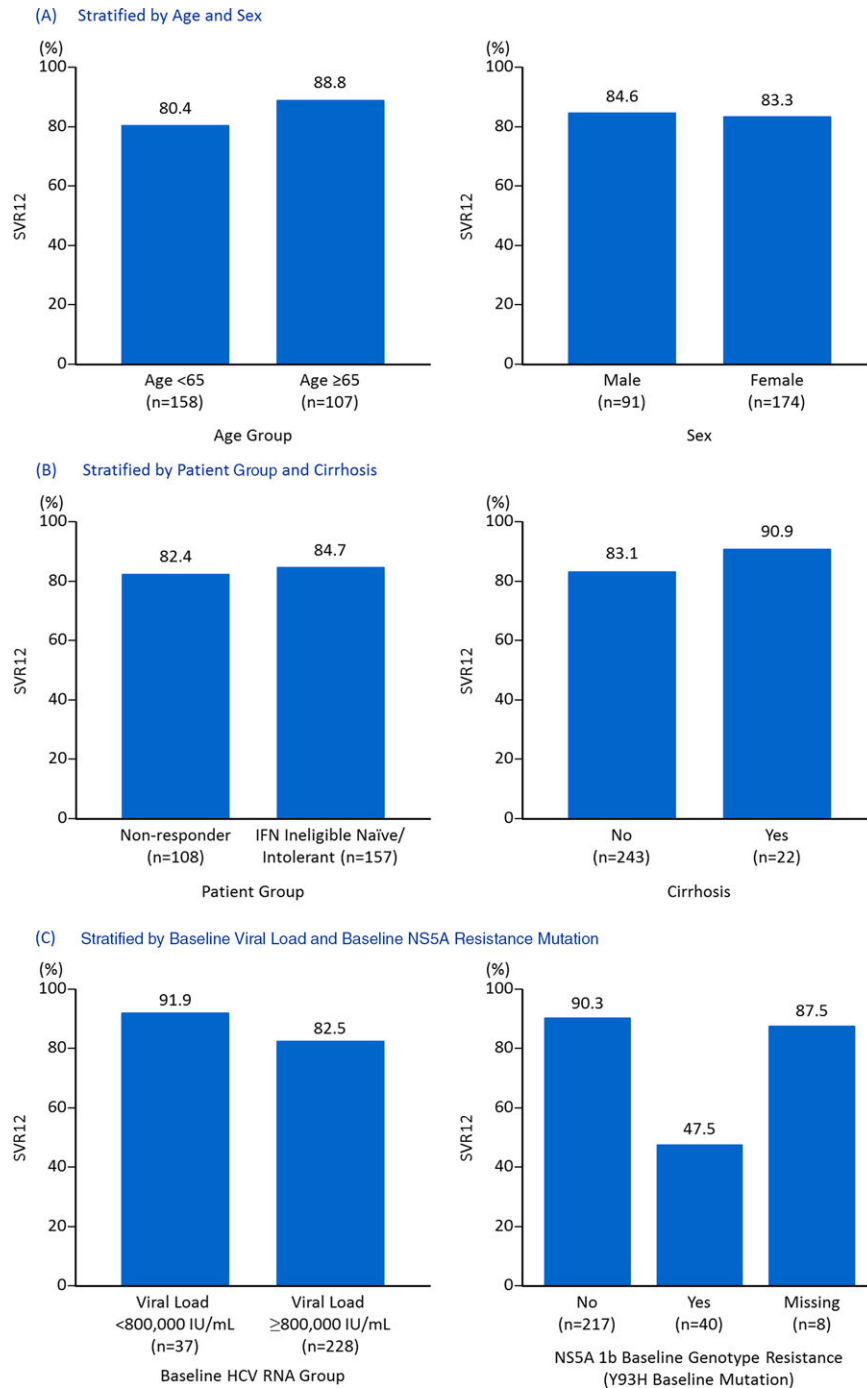


Figure 1. Observed SVR12 rates (A) stratified by sex and age, (B) stratified by patient group and cirrhosis, (C) stratified by baseline viral load and baseline NS5A resistance mutation.

estimates for SVR12 are provided in Table 2. Although baseline Y93H resistance data for 8 patients were missing, model development was performed, coding them as missing. The estimate of Y93H Missing means the effect of missing group relative to patients without Y93H mutation, which is the reference category. Confidence intervals of the final model parameter estimates were obtained by bootstrap. In addition, the exposures

for DCV were identified as a statistically significant predictor of SVR12 (95%CI excludes 0). The odds ratio reflects the increase in odds of achieving SVR12 for patients with Y93H mutation relative to patients who do not have the mutation.

The condition number was calculated as an indicator of the stability of the parameter estimates. The condition number for the final covariate model for SVR12

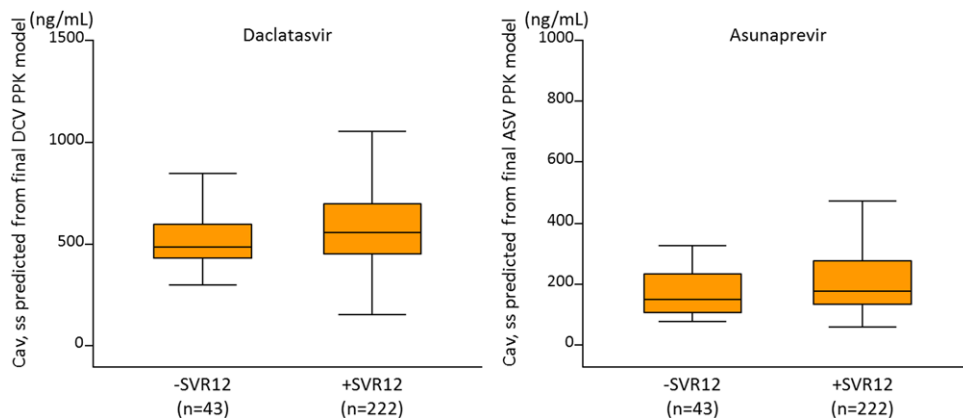


Figure 2. Daclatasvir and asunaprevir average concentrations at steady state in patients with and without SVR12. The lower and upper ends of the boxes represent the 25th and 75th percentiles of the distribution; the line in the box represents the median; and the whiskers are drawn from the upper edge of the box to the largest value within 1.5 times the interquartile range above the 75th percentile and from the lower edge of the box to the smallest value within 1.5 times of the interquartile range below the 25th percentile.

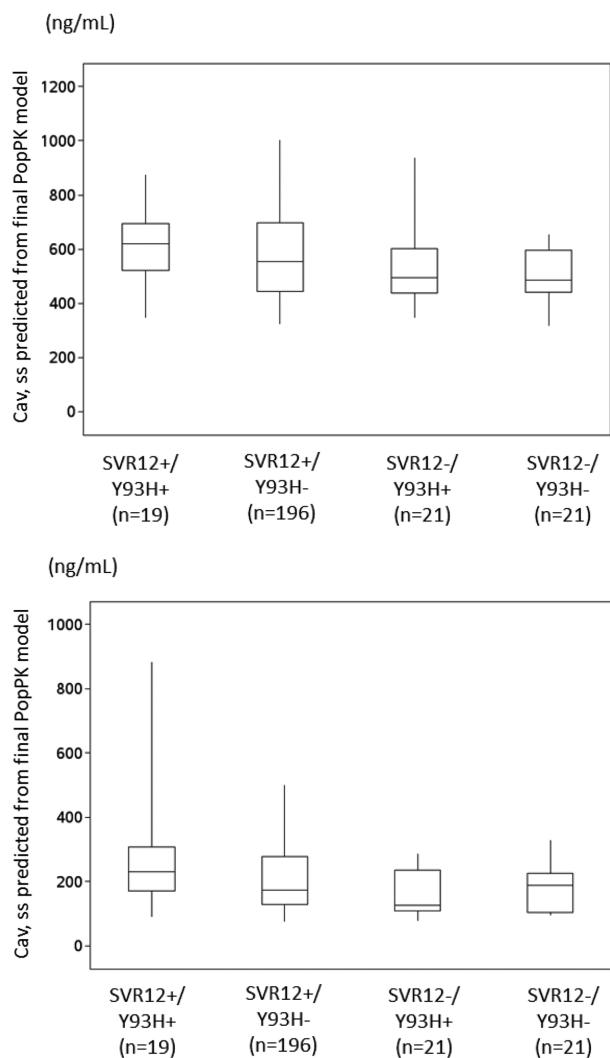


Figure 3. Distribution of $C_{av,ss}$ by the combination of SVR12 and Y93H mutation (upper DCV; lower ASV). The line in the middle of the box is the median; the box is the interquartiles; and the whiskers are the 5th and 95th percentiles.

was 51.5, which is well below the condition number threshold of 1000 that indicates co-linearity and ill conditioning.

Model Evaluation and Model Simulation

Model evaluation was conducted using VPC based on the final model and was presented stratified by Y93H, the sole significant covariate. The results from the predictive check are provided in Figure 4. In these plots, the 95% prediction intervals of the SVR12 response rates were summarized from 1000 trials by each bin, categorized by quartiles of $C_{av,ss}$ and compared with the observed proportion of each bin for $C_{av,ss}$. In addition, the model-predicted probability (95%CI of SVR12 obtained from bootstrap) are presented. The simulation was performed, fixing at median exposure values (ASV $C_{av,ss} = 176$ ng/mL or DCV $C_{av,ss} = 557$ ng/mL). Patients with Y93H mutation had a lower response rate and wider confidence intervals due to a smaller sample size, especially at high exposure. The median predictions followed the trend of the relationships, and most of the observed proportions were covered by the 95% intervals of the model predictions. The plots clearly showed not only the robustness and stability of the model in predicting the SVR12 rates, but the effect of the Y93H mutation. VPC plots showed there was good agreement between the model predicted probability of SVR12 responders and the observed proportion.

Discussion

The exposure-efficacy response analysis was conducted to evaluate the relationship between exposure of ASV and DCV and efficacy measures (SVR12) in the Japanese HCV-infected patients who were either nonresponders to previous IFN/RBV treatment or were ineligible/intolerant to IFN. The analysis was con-

Table 2. Final Model Parameter Estimates for SVR12

Name	Estimate	Standard Error (RSE%) ^a	95%CI ^b
Slope of ASV (β_1)	0.0069	0.00236 (34.2)	-0.00109 to 0.0103
Slope of DCV (β_2)	0.00368	0.000587 (16.0)	0.00192 to 0.00501
Interaction between ASV and DCV(β_3) (β_10^{-3})	-0.00912	0.00253 (27.7)	-0.0122 to 0.0154
Y93H (CAT1)	-2.53	0.415 (16.4)	-3.48 to -1.83
Y93H Missing (CAT2)	-0.508	1.10 (217)	-2.12 to 705

^aRSE% is the relative standard error (standard error as a percentage of estimate).

^bConfidence interval values are taken from bootstrap calculations.

ducted using a logistic regression approach with ASV and DCV $C_{av,ss}$ predicted from the individual popPK analysis as measures of exposure and SVR12, which was the efficacy end point in the clinical studies and the surrogate of cure. A viral kinetic modeling approach would be helpful to understand the dynamic of HCV RNA levels. The approach may enable proposal of a new usage for an antiviral drug (eg, shortened treatment).¹⁶ In this analysis, we selected the E-R approach using an efficacy end point that had been established to show the appropriateness of dose selection of ASV and DCV. Regarding the rationale for selecting $C_{av,ss}$ for assessing association with efficacy, considering the duration of treatment and preferential distribution for the site of activity (liver), especially for ASV, overall exposure throughout a dosing interval as measured by AUC or $C_{av,ss}$ (AUC/dosing interval), rather than plasma concentration at any single time point (including minimum or maximum observed plasma concentration) was likely to correlate best with antiviral response. This is supported by the fact that the transient rise and fall of plasma concentration is likely not reflected as significantly within the liver, where more consistent concentrations would be expected at steady state. Therefore, we analyzed the relationship between ASV and DCV $C_{av,ss}$, (a surrogate for steady-state AUC) and SVR12 response rates. A prespecified model including an intercept term (absence of both drugs), effect of ASV and DCV, and an interaction term between ASV and DCV $C_{av,ss}$ were included in the model.

AI447017 and AI447026 for the DUAL combination in Japanese HCV-infected patients evaluated only 1 dose of DCV (60 mg once daily), but 2 doses of ASV were evaluated in the phase 2 study (200 mg twice daily and 600 mg twice daily of the tablet formulation). Because only 1 dose of DCV was tested in the Japan DUAL program, the effect of DCV was included using a linear linked function model in the E-R analysis. However, although the efficacy rates in AI447017 and AI447026 were relatively similar, based on the wide range of model-predicted ASV exposures across doses in the studies (200-mg twice-daily and 600-mg twice-daily tablet and 100-mg softgel capsule), the effect of

ASV in the E-R logit model was tested using linear and nonlinear relationships.

As described above, a model with linear effect of ASV and DCV with an interaction term between ASV and DCV exposures was identified as the optimal base model describing the relationship between the SVR12 rate and drug exposure. The base model clearly indicated that the SVR12 rate was significantly greater with increasing exposures of ASV and DCV in the therapeutic exposure range, in the absence of other covariates. An interaction term between ASV and DCV exposures was retained in the model based on the significant decrease in objective function value. This means that the E-R relationship of ASV or DCV is influenced by the exposure of the interacting drug. Although the slope for the interaction between ASV and DCV exposures was negative, the impact of the interaction was not expected to be large given the range of ASV and DCV exposures in the current dataset and the relatively high rate of SVR12 achieved for the DUAL treatment.

The base model was utilized to identify significant covariates/predictors for describing SVR12. The pre-specified predictors included demographic covariates such as age, body weight, and sex and disease predictors such as baseline viral load, patient type, baseline ALT, *IL28B* GT, and cirrhosis status. In the final model, only the Y93H mutation for NS5A was identified as a significant covariate. Based on the model estimate, patients with the mutation have a significantly lower SVR rate at similar exposures of ASV and DCV. It has been shown that the Y93H mutation confers a loss in DCV potency (DCV half-maximal effective concentration increased 19-fold over wild-type in transient HCV replication assays).¹⁷ However, as only data for DCV 60 mg once daily were used for the E-R analysis and the number of subjects was not large, it would be difficult to confirm how the changes in response compares to the DCV exposure range versus the fold-shift in half-maximal effective concentration values.

One patient with the Y93F amino acid substitution was confirmed in the phase 3 study, however, the effect of this substitution on virologic response cannot be concluded due to the small sample size. In addition

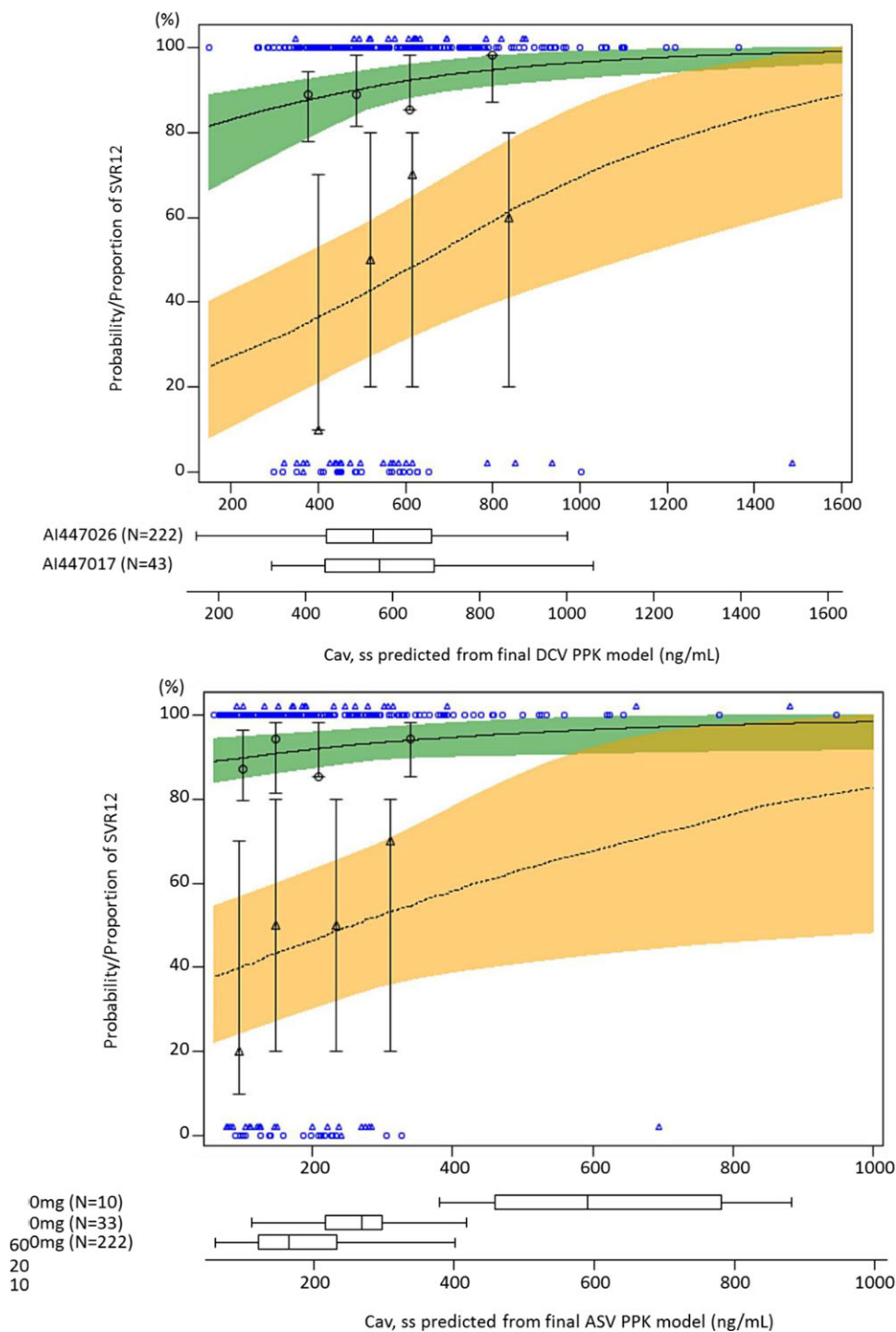


Figure 4. Observed proportion and model predicted probability of SVR12 versus $C_{av,ss}$ and the effect of NS5A Y93H resistance mutations (upper DCV; lower ASV). The symbols represent the proportion of responders, grouped by quartiles of $C_{av,ss}$ and plotted at the median for the groups (circle, patients without Y93H mutation; triangle, patients with Y93H mutation). The centered curves and shaded areas represent median values and 95% CIs of the model-predicted response probability, respectively (solid line, patients without Y93H mutation; dotted line, patients with Y93H mutation). The vertical bars represent the 95% model prediction intervals of the SVR12 rate, grouped by quartiles of $C_{av,ss}$ and plotted at the median for the groups. The box plot shows the distribution of $C_{av,ss}$ by dose or study groups; the left and right ends of the boxes represent the 25th and 75th percentiles of the distribution, the line in the box represents the median, and the whiskers are drawn from the right edge of the box to the largest value within 1.5 times of the interquartile range above the 75th percentile, and from the left edge of the box to the smallest value within 1.5 times of the interquartile range below the 25th percentile.

to the Y93H mutation, the signature NS5A RAVs NS5A-L31M/V also pre-existed in 3.6% (8 of 222) patients in the phase 3 study and 75% of these patients failed to achieve SVR. However, this variant was not included in the E-R analysis as a covariate because the number of patients with the variant was less than the prespecified criteria (covariate present in >5% of patients) for testing covariate effects. Only one patient in the phase 3 study had both L31M/V and Y93H at baseline.

Baseline viral load was thoroughly assessed based on the results of the previous analysis for phase 3 dose selection and the results from the phase 3 study (AI447026). Results from AI447026 indicated that patients with baseline viral load <800,000 IU/mL had a higher rate of SVR24 (93.9%) compared with patients with baseline viral load \geq 800,000 IU/mL (83.1%).⁹ The covariate effect was evaluated as a continuous variable and as a categorical value (baseline viral load <800,000 IU/mL and \geq 800,000 IU/mL). The results indicated that baseline viral load using either method was not a significant covariate at the 1% level of LRT. This could be attributed to the relatively smaller number of patients with baseline viral load <800,000 IU/mL ($n = 33$ vs $n = 189$ for \geq 800,000 IU/mL).

It should be noted that even though the sample size was small ($n = 22$), the population with cirrhosis present at baseline did not predict a lower SVR12 rate. Actually, cirrhosis was not significant in the E-R model and the observed SVR12 rates was 90.9% in the subjects with cirrhosis.

The evaluation for the final model was conducted using a VPC and simulation approach. The simulations were categorized based on the presence or absence of the baseline Y93H mutation. Model evaluation plots shown in Figure 4 demonstrate that the final model was able to predict the observed SVR12 rates (depicted as SVR12 rates for each quartile of exposure). In addition, the model-predicted response rate also show that patients who did not have a preexisting NS5A Y93H mutation had a relatively flat E-R relationship for both ASV and DCV (doubling of median ASV and DCV exposure resulted only in a <10% increase in SVR12 rate). The simulations also clearly demonstrate that at median DCV exposures, there was no clear benefit in virologic response with higher ASV exposures (equivalent to the 600-mg twice-daily tablet) in patients without the Y93H mutation. In patients with the mutation, however, a more sensitive E-R relationship was observed. The simulations indicate that patients with the Y93H mutation at baseline were predicted to have low SVR12 rate at low drug exposures and theoretically could have achieved higher rates of SVR12 if higher exposures were achieved. It should be noted that based on the observed data, 1 of 2 patients with the Y93H mu-

tation at baseline and ASV and DCV $C_{av,ss}$ higher than the 75th percentile achieved SVR12. All 5 patients with the Y93H baseline mutation and ASV and DCV $C_{av,ss}$ lower than the 25th percentile did not achieve SVR12. Although the data trends toward patients with the Y93H mutation at baseline having lower drug exposure, no definitive conclusions regarding the relationship of E-R in patients with Y93H can be made due to the small number of patients with the mutation in the current dataset ($n = 40$ of 265 patients had pre-existing Y93H mutation).

Results from the ASV popPK analysis suggest that the exposure in the patients administered the 600-mg twice-daily tablet of ASV in AI447017 was slightly underpredicted. Therefore, the E-R analysis was also conducted without the 600-mg twice-daily tablet data in the final model to assess the impact of the 600-mg twice-daily cohort on the exposure-efficacy relationship. The results of the analysis after excluding the 600-mg twice-daily cohort are presented in Supplementary Table 4. The E-R model parameter estimates were similar regardless of 600-mg twice-daily data exclusion, suggesting that the final E-R model is robust and can be used for predictions.

Model-based estimates indicate that the rates of SVR12 at median $C_{av,ss}$ for the 100-mg twice-daily softgel capsule for ASV and the 60-mg once-daily tablet for DCV were 91.2% in patients without Y93H baseline mutation. The rate of SVR12 was 45.5% in patients with Y93H baseline mutation, which was consistent with the observed result from clinical studies (47.5%). In addition, the rates of SVR12 at median $C_{av,ss}$ for the 200-mg twice-daily tablet for ASV and 60-mg once-daily tablet for DCV in patients were similar to those of the ASV softgel capsule regimen (92.7% and 50.2% in patients without and with Y93H baseline mutation, respectively).

The exposure-HCV antiviral E-R analyses results demonstrate that for the 100-mg twice-daily softgel ASV capsule and the 60-mg once-daily DCV tablet, robust SVR12 was achieved in Japanese patients infected with HCV GT 1b, particularly in the absence of the Y93H mutation at baseline.

Conclusion

In conclusion, the presence of the signature NS5A Y93H mutation at baseline was the only significant parameter of SVR12 in the final E-R model. There is no evidence of a clinically meaningful effect of the following covariates on SVR12 rate: baseline age, baseline body weight, sex, baseline CrCL, baseline ALT level, *IL28B* GT (rs12979860), baseline viral load, patient type (nonresponder or IFN-based therapy ineligible naive/intolerant patient), cirrhosis (yes or no), study

(AI447017 or AI447026), and OATP1B1 haplotype. Model evaluation plots demonstrated that the final model was able to predict the observed SVR rates. The E-R analyses support the clinical utility of the DUAL regimen of 60-mg once-daily DCV and 100-mg twice-daily ASV in Japanese patients infected with HCV GT 1b.

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Declaration of Conflicting Interests

Takayo Ueno, Mayu Osawa, Hiroki Ishikawa, Yasuhiko Imai, and Tushar Gallimera are employees of Bristol-Myers Squibb K.K./Bristol-Myers Squibb and/or stockholders.

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Supporting Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.