EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

26 June 2014
EMA/419836/2014
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Daklinza

International non-proprietary name: daclatasvir

Procedure No. EMEA/H/C/003768/0000

Note

Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted.

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List of abbreviations

Abbreviation	Term
AE(s)	Adverse event(s)
ALT	Alanine aminotransferase
ANC	Absolute neutrophil count
AST	Aspartate aminotransferase
ASV	Asunaprevir (BMS-650032) NS3 protease inhibitor
AUC	Area under the plasma concentration-time curve
BID	Twice daily
BMI	Body mass index
BMS	Bristol-Myers Squibb
BMS-650032	NS3 protease inhibitor
BMS-790052	NS5A inhibitor
BMS-791325	NS5B polymerase inhibitor
BOC	Boceprevir
CHC	chronic hepatitis C
Cmax	Maximum concentration
CQA	Critical quality attribute
CSR(s)	Clinical study report(s)
CU	Compassionate use
CYP	Cytochrome P450
CYP3A4	Cytochrome P450 3A4
DAA	Direct acting antiviral agent
DAIDS	Division of AIDS
DCV	Daclatasvir (BMS-790052) NS5A inhibitor
DDI	Drug-drug interaction
DOE	Design of experiments
ECG	Electrocardiogram
GC	Gas chromatography
GT(s)	Genotype(s)
GT-1	Genotype 1
GT-1a	Genotype-1a
GT-1b	Genotype-1b
GT-2	Genotype 2
GT-3	Genotype 3
GT-4	Genotype-4
HCC	Hepatocellular carcinoma
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus

Abbreviation	Term
HPLC	High performance liquid chromatography
ICH	International Conference of Harmonization
IFN	Interferon
IFNα	Interferon-alfa
IR	Infrared
KF	Karl Fischer
LLOQ	Lower limit of quantitation
MedDRA	Medical Dictionary of Regulatory Activities
MS	Mass spectra
NS3	Nonstructural protein 3
NS5B	Nonstructural protein 5B
pDILI	Potential drug-induced liver injury
PDR	Protocol defined response
pegIFN	Pegylated interferon
pegIFNα	Pegylated interferon alfa
pegIFNα/RBV	Pegylated interferon alfa plus ribavirin
P-gp	P-glycoprotein
Ph. Eur.	European Pharmacopoeia
PK	Pharmacokinetics
PT	Preferred term
PVC	Polyvinyl Chloride
QbD	Quality by design
QD	Once daily
RBV	Ribavirin
RH	Relative humidity
RNA	Ribonucleic acid
SAE(s)	Serious adverse event(s)
SAP	Statistical analysis plan
SCE	Summary of Clinical Efficacy
SCS	Summary of Clinical Safety
SI	Système International
SmPC	Summary of Product Characteristics
SOC	Standard of care
SOF	Sofosbuvir
SVR	Sustained virologic response
SVR12	Sustained virologic response for 12 weeks after the last dose of study drug
SVR24	Sustained virologic response for 24 weeks after the last dose of study drug
TAMC	Total aerobic microbial count
TD	Target detected
Tmax	Time to maximum concentration

Abbreviation	Term
TND	target not detected
TSE	Transmissible Spongiform Encephalopathy
TVR	telaprevir
TYMC	Total combined yeast and mould count
ULN	Upper limit of normal
UV	Ultraviolet
VBT	Virologic breakthrough
XRD	X-ray diffraction

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Bristol-Myers Squibb Pharma EEIG submitted on 3 December 2013 an application for Marketing Authorisation to the European Medicines Agency (EMA) for Daklinza, through the centralised procedure falling within the Article 3(1) and point 3 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 30 May 2013.

The applicant applied for the following indication: "Daklinza is indicated in combination with other agents for the treatment of chronic hepatitis C virus (HCV) infection in adult patients with compensated liver disease (including cirrhosis).

See section 5.1."

The legal basis for this application refers to:

Article 8.3 of Directive 2001/83/EC - complete and independent application. The applicant indicated that daclatasvir was considered to be a new active substance.

The application submitted is composed of administrative information, complete quality data, non-clinical and clinical data based on applicants' own tests and studies and/or bibliographic literature substituting/supporting certain tests or studies.

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision(s) P/0166/2012 on the agreement of a paediatric investigation plan (PIP).

At the time of submission of the application, the PIP was not yet completed as some measures were deferred.

Information relating to orphan market exclusivity

Similarity

The application did not contain a critical report pursuant to Article 8 of Regulation (EC) No. 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, addressing the possible similarity with authorised orphan medicinal products because there is no authorised orphan medicinal product for the condition related to the proposed indication.

New active Substance status

The applicant requested the active substance daclatasvir contained in the above medicinal product to be considered as a new active substance in itself, as the applicant claims that it is not a constituent of a product previously authorised within the Union.

Scientific Advice

The applicant received Scientific Advice from the CHMP on 18 December 2008 and 18 November 2010. The Scientific Advice pertained to non-clinical and clinical aspects of the dossier.

Licensing status

The product was not licensed in any country at the time of submission of the application.

Manufacturer responsible for batch release

Bristol-Myers Squibb S.r.l.
Loc. Fontana del Ceraso
03012 Anagni (FR)
Italy

1.2. Steps taken for the assessment of the product

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Filip Josephson Co-Rapporteur: Robert James Hemmings

- The application was received by the EMA on 3 December 2013.
- Accelerated Assessment procedure was agreed-upon by CHMP on 21 November 2013.
- The procedure started on 26 December 2013.
- The Rapporteur's first Assessment Report was circulated to all CHMP members on 14 March 2014. The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on 13 March 2014. In accordance with Article 6(3) of Regulation (EC) No 726/2004, the Rapporteur and Co-Rapporteur declared that they had completed their assessment report in less than 80 days.
- During the meeting on 25 April 2014, the CHMP agreed on the consolidated List of Questions to be sent to the applicant. The final consolidated List of Questions was sent to the applicant on 29 April 2014.
- The applicant submitted the responses to the CHMP consolidated List of Questions on 23 May 2014.
- The Rapporteurs circulated the Joint Assessment Report on the applicant's responses to the List of Questions to all CHMP members on 3 June 2014.
- PRAC Risk Management Plan advice and assessment overview was adopted by PRAC on 12 June 2014.
- During the meeting on 26 June 2014, the CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a positive opinion for granting a Marketing Authorisation to Daklinza.

2. Scientific discussion

2.1. Introduction

Hepatitis C virus (HCV) infection is a major European public health challenge, with a prevalence of 0.4-3.5% in different EU member states. It is the most common single cause of liver transplantation in the Union.

HCV is divided into six major genotypes and numerous subtypes, which are based on phylogenetic relationship. Genotype 1 is the most common genotype in Europe, comprising approximately 70 % of infections. Genotype 3 is second most common, followed by genotype 2. Genotype 4 is predominant in Egypt, the nation in the world with the highest documented HCV prevalence. Genotypes 5 and -6 are uncommon in Europe and the US, but are more common in South Africa and South-East Asia, respectively

(Simmonds et al, Hepatology 2005). HCV genotype does not clearly impact the rate of disease progression. Treatment response, however, with available regimens, differs between genotypes.

The goal of antiviral therapy against HCV is to reach sustained virological response (SVR), which is traditionally defined as the absence of quantifiable virus in plasma at least 24 weeks after the end of therapy. However, most relapses occur within 4 weeks of treatment discontinuation, and a 98-99% concordance has been shown between absence of quantifiable virus 12 weeks after therapy, and SVR24 (Florian et al, AASLD 2011). Therefore the absence of measurable virus 12 weeks post end of treatment (SVR12) is presently accepted by European and US regulators as the primary endpoint in clinical trials. Though occasional late relapses occur, in general the durability of SVR has been amply demonstrated (see e.g., Ng and Saab, Clin Gastroenterol Hepatol 2011).

Up until the European commission approval of sofosbuvir (Sovaldi) in early 2014, all approved therapeutic regimens for hepatitis C virus infection contained an interferon. For the treatment of genotype 1 infection, the addition of either one of the NS3/4A protease inhibitors telaprevir or boceprevir approved in 2011, was considered standard-of-care. For genotypes other than -1 there were no direct acting antivirals (DAA) approved, bi-therapy with pegIFN/RBV being the standard. Interferon-based therapies are associated with potentially serious side effects that are important in limiting real life effectiveness. These include a risk of hepatic decompensation and septicaemia in patients with advanced liver disease, as well as bone marrow suppression. Also, there are psychiatric side effects such as depression, which considerably limits eligibility to treatment in the target population (e.g., Bini et al, Am J Gastroenterol 2005).

The approval of sofosbuvir heralded shorter and likely more effective interferon-based therapies for all genotypes. It also made interferon-free treatment options possible. The efficacy of an interferon-free regimen of sofosbuvir+ribavirin, however, is not fully optimised when treating other genotypes than -2; in particular, an increased rate of virological relapse post treatment is anticipated in those patients with most advanced liver disease.

With the approval of NS3/4A inhibitor simeprevir, it is anticipated that a highly effective interferon-free combination regimen with sofosbuvir will be available for more patients. Efficacy in patients with prior exposure to NS3/4A inhibitors telaprevir or boceprevir, however, has not been studied, and could be impaired by prior selection of cross-resistant viral variants.

Thus, despite a very rapid development of new therapies, including interferon-free regimens, there remains an unmet medical need for many European patients with hepatitis C virus infection.

2.2. Quality aspects

2.2.1. Introduction

The finished product is presented as film-coated tablets containing 30 mg or 60 mg of daclatasvir as active substance.

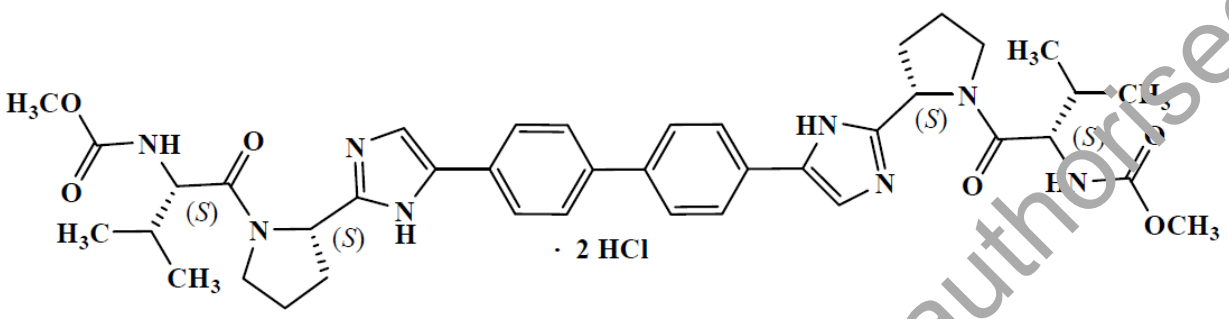
Other ingredients are: anhydrous lactose, microcrystalline cellulose, croscarmellose sodium, silicon dioxide, magnesium stearate, hypromellose, titanium dioxide, macrogol 400, indigo carmine aluminum lake, yellow iron oxide.

The product is available in polyvinyl chloride/poly-chloro-tri-fluoro-ethylene (PVC/PCTFE) clear blister/aluminum foil lidding.

2.2.2. Active Substance

General information

The chemical name of daclatasvir is methyl((1S)-1-(((2S)-2-(5-(4'-(2-((2S)-1-((2S)-2-((methoxycarbonyl)amino)-3-methylbutanoyl)-2-pyrrolidinyl)-1H-imidazol-5-yl)-4-biphenylyl)-1H-imidazol-2-yl)-1-pyrrolidinyl)carbonyl)-2-methylpropyl)carbamate dihydrochloride and has the following structure:



The structure of the active substance has been confirmed by UV, IR, Raman and ^{1}H and ^{13}C NMR spectroscopy, MS spectrometry, and crystal X-Ray diffraction.

Daclatasvir is a white to yellow crystalline non-hygroscopic powder. It is freely soluble in water, dimethyl sulfoxide, methanol; soluble in ethanol (95%); practically insoluble in dichloromethane, tetrahydrofuran, acetonitrile, acetone and ethyl acetate.

Daclatasvir is a chiral molecule with four stereocenters (1,1', 2, 2;) in the S configuration. The synthetic strategy and process design such as starting material and reagent selection, process parameters, and in-process controls ensure the desired configuration at each of the four chiral centers. In addition, the established control strategy minimizes epimerization and eliminates other diastereomeric impurity formation in each step.

Polymorphism has been observed for daclatasvir hydrochloride. Although two neat crystalline dihydrochloride salts, N1 and N-2 have been identified in screening studies, it has been confirmed that the form N-2 is the thermodynamically most stable polymorph and only this form produced by the proposed synthetic process.

Manufacture, characterisation and process controls

Daclatasvir dihydrochloride is synthesised in three main steps using three commercially available well defined starting materials with acceptable specifications. The synthesis involves an alkylation and formation of the imidazole ring, a coupling reaction and the formation of the hydrochloride salt.

As mentioned above, the synthetic process has been designed to ensure the correct configuration at each of the four chiral centres is achieved. In addition, it has been demonstrated that the stereogenic centres do not epimerize during normal or stressed processing conditions.

The manufacturing process has been developed using a combination of conventional univariate studies and elements of QbD such as risk assessment.

The characterisation of the active substance and its impurities are in accordance with the EU guideline on chemistry of new active substances. Potential and actual impurities were well discussed with regards to their origin and characterised.

Adequate in-process controls are applied during the synthesis. The specifications and control methods for intermediate products, starting materials and reagents have been presented.

Specification

The active substance specification includes tests for: appearance, colour, identity (IR/Raman, HPLC), assay (HPLC), impurities (HPLC), residual solvents (GC), HCl content (titration), total inorganic impurities (ICP-MS), and particle size (laser light scattering). The absence of a test for chiral purity in the active substance specification has been adequately justified based on the stereochemical control during the synthetic process and demonstration that there is no epimerization during normal or stressed processing conditions. Similarly, since the N-2 form of daclatasvir hydrochloride is the thermodynamically most stable polymorph and, is consistently produced by the synthetic process and remained unchanged during storage under long-term or accelerated conditions, this parameter is not included in the specification.

The analytical methods used have been adequately described and non-compendial methods appropriately validated in accordance with the ICH guidelines.

Batch analysis data on eleven commercial scale batches of the active substance have been provided. The results are within the specifications and consistent from batch to batch.

Stability

Stability data were provided on three pilot scale batches of active substance manufactured by the proposed commercial manufacturing process stored in a container closure system representative of that intended for the market. Studies were carried out, according to the ICH guidelines, under long term conditions at 5°C/60% RH, 25 ºC/60% RH (18 months) and 30 ºC/65% RH (12 months) and under accelerated conditions at 40 ºC / 75% RH (6 months). Photostability testing following the ICH guideline Q1B was performed on one batch. Results on stress conditions in aqueous solution under acidic (HCl 0. 1N), basic (0.01 N NaOH) and oxidative (0.3 % hydrogen peroxide) conditions; and solid state: heat and humidity (80°C/75% RH) and heat (80°C) were also provided on one batch.

The following parameters were tested: colour and appearance, identification (Raman), assay (HPLC), impurities (HPLC), water content (KF), X-ray diffraction (XRD). The analytical methods used were the same as for release, with the addition of X-ray powder diffraction and water content (KF).

The stability results showed little to no change in colour and appearance, assay, impurities, or X-ray diffraction. A slight increase in the mean water content was observed, but the results were within the predefined specification at all time points.

The results from the forced degradation studies showed that daclatasvir hydrochloride is susceptible to degradation in solution at basic conditions and at high intensity UV and visible light. Minor degradation is observed under oxidative conditions. None of the degradants from the forced degradation studies were observed during the accelerated or long term stability studies.

The stability results indicate that the drug substance manufactured by the proposed supplier is sufficiently stable. The stability results justify the proposed retest period in the proposed container.

2.2.3. Finished Medicinal Product

Description of the product and Pharmaceutical development

The aim of the pharmaceutical development was to obtain immediate release film-coated tablets containing 30 mg or 60 mg of daclatasvir for oral administration to adult patients, which meet compendial and other relevant quality standards, and have a shelf life of at least 2 years.

During the development the relevant physicochemical and biological properties of the drug substance that could influence the performance of the drug product and its manufacturability were studied. These included: polymorphic form, particle size and impurity level.

The critical quality attributes (CQAs) that can impact the safety and efficacy of daclatasvir hydrochloride tablets are: appearance, assay (potency), impurities, content uniformity and dissolution.

The formulation and manufacturing process development have been evaluated through the use of risk assessment and design of experiments (DOE) in order to establish linkages between inputs (raw materials, process parameters), intermediate attributes, and critical quality attributes (CQAs). Extensive development studies have been carried out in order to acquire better understanding of the manufacturing process and to define appropriate control strategy to produce a consistent quality product.

A drug-excipient compatibility study was conducted to screen potential excipients to be used in the formulation. The results from this study showed that microcrystalline cellulose, anhydrous lactose, croscarmellose sodium, silicon dioxide, and magnesium stearate are compatible with the drug substance under dry conditions, and were found to be acceptable for use in the daclatasvir dihydrochloride tablets. All the excipients used are well known pharmaceutical ingredients and their quality is compliant with Ph. Eur standards. There are no novel excipients used in the finished product formulation. The list of excipients is included in section 6.1 of the SmPC.

For Phase 2 clinical studies three strengths (3 mg, 10 mg, and 30 mg) of immediate release film-coated tablets were developed using a roller compaction (dry granulation process). For Phase 3 clinical trials two strengths (30 mg and 60 mg) of immediate release film-coated tablets were used. Several changes were made from the Phase 2 to the Phase 3 formulation, including: an increase in drug loading from 10% w/w to 22% w/w to maintain an acceptable tablet size for the required strengths, optimization of the levels of some of the excipients, and change of the tablet shape. The formulation composition and dry granulation process for the phase 3 and commercial tablets are identical.

A relative bioavailability study to compare the phase 3 tablet formulation (1 x 60 mg) against the phase 2 tablet formulation (2 x 30mg) was conducted. The results from this study showed that comparable systemic exposure to daclatasvir was achieved with both formulations. The formulation changes from the Phase 2 to the Phase 3 tablets were also assessed using in vitro dissolution testing to support the use of tablet multiples and transition to the Phase 3 formulation. These studies showed equivalent dissolution of the 2 x 30 mg Phase 2 tablets to 2 x 30 mg and 1 x 60 mg Phase 3 tablets. The discriminatory power of the dissolution method used to bridge between phase 2 and phase 3 formulations and proposed for quality control was also adequately demonstrated.

The primary packaging is polyvinyl chloride/poly-chloro-tri-fluoro-ethylene (PVC/PCTFE/Alu) clear blister/aluminum foil lidding. The material complies with Ph.Eur. and EC requirements. The choice of the container closure system has been validated by stability data and is adequate for the intended use of the product.

Manufacture of the product and process controls

The manufacturing process is a dry granulation process that is applicable to both tablets strengths and includes the following unit operations: pre-blending, roller compaction, final blending (lubrication), tablet compression, film coating and packaging. The process is considered to be a standard manufacturing process.

Major steps of the manufacturing process have been validated by a number of studies. It has been demonstrated that the manufacturing process is capable of producing the finished product of intended quality in a reproducible manner. The in-process controls are adequate for this pharmaceutical form.

Product specification

The finished product release specifications include appropriate tests for this type of dosage form: description, identification (HPLC, IR-ATR), assay (HPLC), impurities (HPLC), dissolution (Ph. Eur.),

content uniformity (Ph. Eur.), microbial limits (TAMC, TYMC and E. coli) (Ph. Eur). The absence of a test for water content has been adequately justified.

Batch analysis results are provided for 4 pilot scale batches and 5 commercial scale batch of the 30 mg tablets, and 7 pilot scale batches and 7 commercial scale batches of the 60 mg tablets confirming the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

The finished product is released onto the market based on the above release specifications, through traditional final product release testing.

Stability of the product

Stability data on three pilot scale batches of 30 mg and 60 mg film-coated tablets stored under long term conditions for 18 months at 5 ºC , 25 ºC / 60% RH and, 30 ºC / 75 % RH; and for up to 6 months under accelerated conditions at 40 ºC / 75% RH according to the ICH guidelines were provided. The batches are representative of those proposed for marketing and were packed in the primary packaging proposed for marketing.

Samples were tested for appearance, identification, potency, impurities, content uniformity, dissolution and water content. The analytical procedures used are stability indicating.

In addition, one batch of each strength was exposed to light as defined in the ICH Guideline on Photostability Testing of New Drug Substances and Products.

The stability data from the long term conditions indicate that daclatasvir dihydrochloride film-coated tablets, 30 mg and 60 mg, are stable through 18 months of storage. The results showed little to no change in all tested parameters.

The stability data from the accelerated condition of 40 ºC / 75% RH indicate that there was essentially no change in the tested parameters during the 6 month study period.

A slight increase in mean water content values was observed in 30 mg and 60 mg tablets stored at the higher humidity conditions of 30 ºC / 75 % RH or 40 ºC / 75% RH, and 60 mg tablets stored at 25 ºC / 60% RH, but these increases had no impact on other attributes.

Open dish studies and the photostability study conducted indicate that the tablets are not sensitive to moisture or light.

Based on available stability data, the shelf-life and storage conditions as stated in the SmPC are acceptable.

Adventitious agents

It is confirmed that the lactose is produced from milk from healthy animals in the same condition as those used to collect milk for human consumption and that the lactose has been prepared without the use of ruminant material other than calf rennet according to the Note for Guidance on Minimising the Risk of Transmitting Animal Spongiform Encephalopathy Agents Via Human and veterinary medicinal products.

No other excipients derived from animal or human origin have been used.

2.2.4. Discussion on chemical, pharmaceutical and biological aspects

Information on development, manufacture and control of the active substance and finished product has been presented in a satisfactory manner. The results of tests carried out indicate consistency and uniformity of important product quality characteristics, and these in turn lead to the conclusion that the product should have a satisfactory and uniform performance in clinical use.

The applicant has applied QbD principles in the development of the active substance and finished product and its manufacturing process, but no design spaces were claimed.

2.2.5. Conclusions on the chemical, pharmaceutical and biological aspects

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SmPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. Data has been presented to give reassurance on viral/TSE safety.

2.3. Non-clinical aspects

2.3.1. Introduction

2.3.2. Pharmacology

Daclatasvir is a first in class direct acting antiviral agent, intended for treatment of hepatitis C virus infection. Daclatasvir binds to and inhibits the function of the hepatitis C virus protein NS5A. NS5A is involved in both viral RNA replication and virus particle assembly. A putative inhibitor-binding region spanning amino acids 21 to 30 of NS5A was identified. Concerning the further primary pharmacology of daclatasvir, see section on pharmacodynamics.

At 10 µM, daclatasvir showed a 65% inhibition binding to the sodium ion channel, but did not display a greater than 50% inhibition or induction of any target in a 36 target assay including standard receptors, enzymes and ion channels or in an assay including receptors for rat aldosteronе, human angiotensin, atrial natriuretic factor and vasopressin. Metabolite BMS 805215, at10 µM, did not show any significant effects on the 37 targets assay.

In a cytotoxicity assay, CC50 values ranging from 17 to 90 µM in liver, kidney, kidney, lung fibroblast cells and lymphocytes were seen. No significant toxicity was observed with daclatasvir treatment for any of the cell types tested.

Cardiovascular effects of daclatasvir and metabolites were evaluated in vitro and in vivo.

In the hERG/IKr assay, the IC50 of dalactasvir was 21.6 µg/mL (29.2 µM), to be compared with 1.73 µg/mL, the highest plasma concentration value for daclatasvir at the maximum recommended human dose. Daclatasvir showed a moderate inhibition of sodium and L-type calcium currents, 32% and 60%, respectively, at 7.39 µg/mL. No effects on Purkinje fibre action potential parameters were observed at the concentrations tested, 2.22, 7.39 and 22.2 µg/mL. However, participation of the drug substance was seen at the two highest concentrations, which questions the usefulness of these studies. Human metabolite BMS-795853 displayed comparable inhibition of cardiac hERG/IKr currents (7.7%, 19.9%, and 40.0% at 0.68, 2.04 and 6.81 µg/mL, respectively), but less potent inhibition of cardiac sodium currents, approximately 22% at 6.81 µg/mL.

In rabbit, administered 1, 3, 10 and 30 mg/kg as an intravenous single dose, moderately increased QRS duration (29 ±1%) and mildly increased PR (19 ±3%), AH (16 ±4%) and HV (10 ±1%) intervals were observed at 30 mg/kg. No atrioventricular conduction block or other cardiac arrhythmias or effects on either QTcf or QTcv intervals were observed. The no effect level, 10 mg/kg, corresponded to a plasma concentration of 72.9 µg/mL. This yields an exposure margin to plasma levels at maximum recommended human dose of approximately 40.

In telemetered dogs, administered 15 or 100 mg/kg as a single oral dose, the highest dose was associated with a reversible moderate increased systolic, diastolic and mean arterial pressure in 4 out of 6 dogs. A 10

to 15% decrease in a calculated index of cardiac contractility was also seen in 4 dogs. The no effect level for cardiovascular effects was 15 mg/kg, which corresponds to a Cmax of 2-4 µg/mL. This is approximately the same plasma level as the clinical levels reached at maximum recommended human dose.

Clinically, the effect of daclatasvir on cardiovascular safety was evaluated in a thorough QT study. Single doses of 60 mg or 180 mg did not have a relevant effect on QTc interval and there was no significant relationship between increased daclatasvir plasma concentration and change in QTc. Daclatasvir does not appear to have a potential for adverse QT-effects.

The safety aspects of the central nervous and respiratory system were not studied in dedicated safety pharmacology studies, but claimed to be evaluated in single and repeat-dose toxicity studies. According to the applicant, no effects were seen on respiratory or central nervous system parameters after oral administration; In mice (up to 1000 mg/kg as a single dose and 100 mg/kg as repeated dosing), rats (up to 1000 mg/kg as a single dose and 50 mg/kg as repeated dosing), dogs (up to 150 mg/kg as a single dose and 50 mg/kg as repeated dosing) and monkeys (up to 300 mg/kg as repeated dosing). However, in the study reports it is not clear how the safety pharmacology aspects of the central nervous system and respiratory system were studied. On the contrary, in several studies (DS07063, DS07054, DS06211, DS07186, DS07055, DS08002, DS07058, DS07214, DS08039 and DS08003), relevant parameters do not seem to have been studied at all. Considering the clinical experience with daclatasvir, the non-clinical data and safety pharmacology assessment of the nervous and respiratory system is considered superseded by clinical data.

2.3.3. Pharmacokinetics

The absorption, distribution, metabolism and excretion of daclatasvir were evaluated in a series of in vitro and in vivo studies conducted in mice, rats, rabbits, dogs and monkeys. In addition, pharmacokinetic/toxicokinetic data were generated in support of toxicology studies.

Absorption: In artificial membrane permeability assays in vitro, the permeability coefficient of daclatasvir was reported to be comparable to that of compounds exhibiting good absorption in humans. The absorption of orally administered daclatasvir in mice, rats, dogs, and monkeys was rapid, with Tmax values being up to 2.0 hours. The absolute bioavailability of daclatasvir was high in mice and dogs but lower in rats and monkeys. In humans oral absolute bioavailability was shown to be 67%. In rats, pharmacokinetic data suggested that the oral bioavailability is unlikely to be limited by first-pass hepatic clearance. There was evidence indicating that in dogs the oral absorption of daclatasvir was pH dependent and that in mice Pgp plays a role in the elimination of daclatasvir. In studies in P-gp-knock-out mice, there was evidence to suggest that P-gp plays a role in the elimination of daclatasvir.

Distribution: In vitro, protein binding of daclatasvir at 10 µM was similar in mouse, rat, rabbit, dog, monkey and human serum, ranging from 95.1% to 99.5%.

Daclatasvir shows covalent binding in liver microsomes. In addition, persistent radioactivity was seen in some tissues in the non-clinical studies. However, there was no evidence of persistent radioactivity in a human ADME study. During the assessment, the applicant provided a discussion about the covalent binding of daclatasvir in liver microsomes and a potential relation to toxicity findings. In conclusion, the risk for potential reactive metabolite-mediated liver toxicity in humans appears low, and hepatotoxicity is included as an important potential risk in the RMP. A risk for potential idiosyncratic reactions is not possible to dismiss based on available data.

There was evidence to suggest that in the blood, the compound is distributed preferentially into plasma.

In rats, dogs, and monkeys following intravenous doses of up to 5 mg/kg, daclatasvir steady state volume of distribution levels were greater than the reported total body water volumes in these species, indicating extravascular distribution.

In the pigmented rats administered 10 mg/kg [14C]-daclatasvir, drug-derived radioactivity was rapidly absorbed and widely distributed. Concentrations of radioactivity were highest in the adrenal gland, bile, liver, caecum, small intestine, and stomach. Similar effects were seen in the non-pigmented rats. However, in pigmented skin and eye uveal tract, the elimination of radioactivity was slower than from non-pigmented tissue. These data suggest a specific, but reversible binding of [14C]-daclatasvir-derived radioactivity to melanin containing tissues.

In a repeat-dose study in non-pigmented rats orally administered [14C]-daclatasvir for 14 days, accumulation of radioactivity was not observed in any tissue and the elimination of radioactivity from most tissues were similar to those observed in the single dose study. At 84 days post dose, most of the tissues were devoid of radioactivity with the exception of exorbital lacrimal gland, intraorbital lacrimal gland, thymus, and thyroid tissues.

In further studies conducted with daclatasvir, the liver-to-serum or -plasma AUC ratios were 2.35 and 1.9 (IV and oral, respectively) in mice, 5.9 and 6.8 (IV and oral, respectively) in rats, 10.6 (oral) in dogs, 17 in monkeys (oral) and > 1 in other tissues. Results from studies in P-gp knock-out mice, suggested that P-gp plays a role in limiting the distribution of daclatasvir into mouse brain.

In pregnant rats administered a single oral dose of [14C]-daclatasvir, the distribution of radioactivity into maternal tissues was similar to non-pregnant rats. Radioactivity was detected in fetal liver, indicating that daclatasvir and/or its metabolites crossed the placenta. In lactating rats administered a single oral dose of [14C]-daclatasvir, drug-related radioactivity was detected in milk with concentrations 1.7- to 2 fold maternal plasma levels.

Metabolism: Metabolism of daclatasvir was qualitatively similar in the toxicology species and humans. The in vivo biotransformation of daclatasvir was characterised by the production of numerous oxidative metabolites, with the number of characterised metabolites detected in excreta ranging from 8 in humans to 16 in mice, rats, rabbits, dogs, and monkeys. In vitro and in vivo the prominent metabolic pathways included oxidative pyrrolidine ring opening followed by intramolecular cyclization (to BMS 805215), carbamate cleavage (to BMS-795853), and other hydroxylated metabolites. CYP3A4 was the primary enzyme involved in the metabolism of daclatasvir.

Daclatasvir was the predominant radioactive component in plasma in animals (75% to 94%) and in humans (97% to 100%). The metabolite BMS-805215 was the only metabolite detected in human plasma representing a minor circulating metabolite with a BMS-805215-to-daclatasvir AUC ratio of ≤5%. BMS-805215 was the major plasma metabolite in monkeys but was minor in mice, rats, rabbits, and dogs. Based on exposure data of daclatasvir in animals (rats, rabbits, dogs and monkeys), BMS-805215 was adequately assessed.

The predominant metabolites identified in human feces were BMS-805215 (15.2% of the dose) and BMS-795853 (4% of the dose). BMS-805215 was detected in intact and bile-duct cannulated monkeys (12.6% and 17.5% of the dose, respectively) and rats (10.5% of the dose). BMS-795853 was detected in mice (6.3% of the dose) and bile-duct cannulated dogs (6% of the dose). Other metabolites identified in bile, feces, or urine represented < 5% of the dose. Overall, the percent of the daclatasvir dose recovered as metabolites was similar in animals and humans.

Dogs were considered an outlier species based on their in vivo metabolic profile. Therefore, monkeys were selected as the non-rodent toxicology species.

Excretion: The elimination of daclatasvir in animals involved multiple pathways including fecal excretion, direct intestinal secretion, and metabolism followed by biliary excretion. Renal clearance was a minor route of elimination for daclatasvir.

Fecal excretion of daclatasvir was higher in humans (52.5% of the dose) than in animals (34%, 24.5%, 51.9%, and 32.3% of the dose in mice, rats, rabbits, and monkeys, respectively). Metabolic clearance of daclatasvir was similar in humans (30.1% of the dose) and animals (19.2% to 27.5% of the dose). Biliary clearance was also an important elimination pathway of daclatasvir and metabolites in animals; a considerable portion of the dose was excreted as daclatasvir (11.5%, 12.5%, and 1.4% of the dose) and metabolites (21.8%, 8%, and 11.7% of the dose) in bile of bile-duct cannulated rat, dog, and monkey, respectively. Since daclatasvir was detected in the bile of rat, dog, and monkey, there may be biliary excretion of daclatasvir in humans.

After intravenous administration to bile-duct cannlulated rats, dogs, and monkeys, 27.2%, 8.05%, and 1.9%, respectively, of the administered dose, was excreted as unchanged daclatasvir in feces, suggesting direct intestinal secretion of daclatasvir possibly due to P-gp or other transporter activity. Therefore, daclatasvir in feces after oral dosing could be due to biliary secretion and intestinal secretion, as well as incomplete absorption. The fraction of an oral dose recovered in urine as unchanged daclatasvir was 0.73% to 1.55% in animals and 6.61% in humans, indicating that renal clearance was a minor pathway of daclatasvir elimination.

Drug interaction: Drug-drug-interactions are presented and discussed in the clinical section of the report.

2.3.4. Toxicology

The toxicological profile of daclatasvir has been evaluated in a comprehensive set of non-clinical studies including single- and repeat-dose toxicity studies in mice, rats, dogs, and monkeys; repeat-dose toxicity studies ≤ 6 months in rats, 1 month in dogs, and ≤ 9 months in monkeys; combination (daclatasvir and pegIFNα/ribavirin) toxicity study (monkey 14 days); genotoxicity; phototoxicity studies; fertility and pre- and postnatal development (rat) and embryo-foetal development (rat and rabbit) studies; juvenile toxicity studies (rat); local tolerance (mouse, rabbit, bovine); and carcinogenicity studies (Tg-rasH2 mice, Sprague-Dawley rats). Immunotoxicity was evaluated by addition of selected immunotoxicity endpoints in dog and monkey repeat dose studies.

The rat (Sprague-Dawley) and monkey (Cynomolgus) were selected as the main rodent and non-rodent toxicology species. In general, the non-clinical toxicology program has been performed according to relevant guidelines.

Single dose toxicity

The single-dose toxicity of daclatasvir is considered low. Single oral doses of ≤150 mg/kg in dogs and monkeys and ≤1000 mg/kg in mice and rats produced no mortality and were well tolerated.

Repeat dose toxicity

Daclatasvir has been tested in repeat-dose toxicity studies in Sprague Dawley rats (up to 6 months with 2 months recovery), Beagle dogs (up to 1 month with 1 month recovery) and Cynomolgus monkeys (up to 9 months with 2 months recovery). No apparent dose-limiting effects were noted in rat and monkey studies and potentially higher dose levels could have been employed. The main target organs that were consistent across 2 or more species included the adrenal gland and liver. Other daclatasvir-related effects noted in bone marrow (dogs and monkeys) and prostate and/or testes (dogs and rats) occurred with either minimal severity or were associated with overtly toxic doses. Most changes were reversible but the mechanisms of toxicity are not known. Pre-terminal mortalities occurred in dogs (10.5-fold clinical exposure based on AUC) and in monkeys (5.3-fold clinical exposure based on AUC).

Pre-terminal mortalities

In the 1-month dog study, a dose of 100 mg/kg/day was associated with pre-terminal euthanatisation of 3 dogs due to liver and/or bone marrow toxicity (see under Liver and Bone marrow) and 1 dog due to an interdigital cyst that had progressed to an abscess and draining fistula. Although the cyst was considered incidental, the progression of the condition is considered likely related to daclatasvir treatment. Additional findings in these pre-terminal dog included effects in spleen/thymus (extramedullary hematopoiesis and/or lymphoid depletion), in pancreas (acinar cell vacuolation), and in male reproductive organs (see under prostate and/or testes). In the 4 decedent dogs, the approximate plasma Cmax at necropsy were higher (23.9 to 32.2 µg/mL) and the daclatasvir liver-to-plasma ratio was lower (< 1) than in high-dose dogs that survived until scheduled necropsy (Cmax ≤9.3 µg/mL, daclatasvir liver-to-plasma ratio > 5). As daclatasvir elimination involves multiple pathways the lower liver-to-plasma ratios may suggest saturated elimination at high exposures.

In the 9-month monkey study, a single monkey given 150 mg/kg/day was euthanatized on Day 28 due to a deteriorating condition attributed to inflammatory changes in several tissues. Minimal to moderate chronic inflammation was noted in liver, lymph nodes, spleen, thymus, kidney, heart and pancreas. Findings in the skin involved both mild to severe epidermal necrosis and ulceration accompanied by inflammation and crust formation. Other findings included infarcts in spleen and stomach with associated ulceration and were considered likely due to coagulopathy. In addition, a marked decreased cytoplasmic cortical vacuolation in adrenal glands was observed. Since an infectious process was suspected, this monkey was given antibiotics and anti-inflammatory agents that confounded a diagnosis and the primary cause for the moribund condition could not be determined. Although most findings in this early decedent were inconsistent with those in all other monkeys given 150 mg/kg/day for an additional 8 months and with monkeys given 300 mg/kg/day for 4 months, a relationship to daclatasvir cannot be excluded. According to the veterinarian, the data is most consistent with a generalised inflammatory process such as septicemia or an idiosyncratic drug reaction.

Adrenal gland

The adrenal gland was a target organ in repeat-dose studies in rats and monkeys. Adrenal gland findings included increases in adrenal gland size/weight, hypertrophy and/or hyperplasia of cortical cells in the zona fasciculata and/or zona reticularis, increases in urine corticosterone levels (rats, at some time-points), and changes in cytoplasmic vacuolation. In monkeys, there were minimal to marked decreases in cytoplasmic vacuolation. Further, adrenal cortical hyperplasia in monkeys was slight and noted in some animals at 300 mg/kg/day (2.7-fold clinical exposure based on AUC) in the 4-month study but was not observed at the highest dose of 150 mg/kg/day (2.6-fold clinical exposure based on AUC) in the 9-month study, despite comparable exposures. All findings were reversible and did not show apparent progression with time. There are no exposure margins to the observed adrenal gland effects and according to the Applicant, the effects are potentially ascribed to stress with limited clinical relevance. Although it is agreed that signs of stress were observed in many studies, histopathological adrenal gland effects were also observed in studies where evidences of stress were not compelling. For example, thymus weight which is a sensitive stress parameter was not generally affected by daclatasvir treatment. Adrenocortical hypertrophy/hyperplasia in the absence of any other stress-associated changes suggests a possible primary effect on adrenocortical hormone synthesis. It was also noted that adrenal gland was one of the organs with the highest [14C]-daclatasvir-derived radioactivity detected for up to 35 days post-dose in the rat QWBA study.

No plausible mechanisms for the observed adrenal gland toxicity, apart from adaptive changes due to stress, is discussed by the applicant. While it is agreed that signs of stress was observed in many studies (deteriorating condition, body weigh effects, food consumption, etc), histopathological adrenal gland effects were also observed in studies where evidences of stress were not compelling (6 months rat study

and 4/9 months monkey studies). For example, thymus weight which is a sensitive stress parameter was not affected by the daclatasvir treatment in any of these studies. Therefore, the evidences of stress are not compelling.

It is agreed that the adrenal gland findings are non-progressive and reversible in all species examined. In clinical trials, adrenal insufficiency was monitored by measurement of 24-hour urine cortisol levels. There were no clinically relevant mean changes over time in 24-hour urine cortisol in three clinical studies (global Phase 1, Japanese Phase 1 and Phase 3 studies). In addition, no trends in clinical parameters consistent with adrenal gland hypertrophy and hypercortisism such as increases in blood pressure and serum glucose levels were observed.

In conclusion, the adrenal gland changes observed in non-clinical studies are likely of low relevance for the human situation.

Liver

Daclatasvir was associated with findings in the liver of rats, dogs, and monkeys. In the rat 1-month study, minimal and reversible hepatic changes including slight increases in serum ALT levels and a minimal increase in liver weights without any histological findings at 100 mg/kg/day for 1 month (7.1-fold clinical exposure based on AUC). These effects did not occur in the 6-month study at ≤ 3.9-fold clinical exposure based on AUC.

In the 9-month monkey study, minimal to moderate Kupffer-cell hypertrophy/hyperplasia correlating with pale foci observed macroscopically in some animals was accompanied by minimal to slight mononuclear cell infiltrates were observed at the high dose of 150 mg/kg/day (2.6-fold clinical exposure based on AUC). Additional findings included increases in ALT, AST and CRP, slight bile-duct hyperplasia in 3 animals, and moderate hepatocellular vacuolation at low incidence. At 30 mg/kg/day (0.8-fold clinical exposure based on AUC), only minimal Kupffer-cell hypertrophy/hyperplasia were observed. There were no liver effects at 15 mg/kg/day (0.2-fold clinical exposure based on AUC). All changes were reversible during the 2-month recovery period, with exception of the minimal to slight Kupffer-cell hypertrophy/hyperplasia in the liver of 1 animal at 30 mg/kg/day and all monkeys at 150 mg/kg/day. In the 4-month monkey study, similar hepatic findings occurred at comparable AUC values (1.5 to 2.7-fold clinical exposures based on AUC).

In the 1-month dog study, a dose of 100 mg/kg/day was associated with mortality of 3 dogs due to liver and/or bone marrow toxicity. The liver findings in the preterminal dogs and those that survived to scheduled necropsy included minimal to moderate perivascular inflammation accompanied by minimal to mild hepatocellular degeneration. Secondary to inflammation and hepatocyte degeneration, there was slight or mild Kupffer-cell hypertrophy/hyperplasia, slight Kupffer-cell pigmentation, and slight sinusoidal neutrophilia. Additionally, there was slight to mild sinus congestion and minimal to mild neutrophil infiltration in the splenic (pancreatic) lymph nodes in two early decedent dogs. Since these lymph nodes drain the liver, the lymph node changes were considered secondary to the liver inflammation. The liver findings correlated with mild to marked increases in bilirubin, ALP and GGT, and slight to moderate increases in ALT and AST. In addition, mild to moderate increases in fibrinogen consistent with an acute phase inflammatory response was closely correlated in occurrence in all dogs, and in severity in most dogs with hepatic perivascular inflammation. The reversibility of the liver findings at the high dose was not evaluated due to loss of animals. At 15 mg/kg/day (1.7-fold clinical exposure based on AUC), minimal perivascular inflammation, slight Kupffer-cell hypertrophy/hyperplasia and pigmentation (1 female); and slight sinusoidal neutrophilia (1 male) were observed. Perivascular inflammation was characterised by accumulations of macrophages and generally fewer neutrophils around central veins and, less commonly, around portal tracts. All these findings were reversible. There were no liver effects at 3 mg/kg/day (0.1-fold clinical exposure based on AUC).

No clinically relevant trends in liver function test were observed in long-term clinical studies when daclatasvir was administered with sufosbuvir or with pegIFNα/ribavirin. Hepatotoxicity is included as an important potential risk in the RMP.

Bone marrow

Bone marrow was a target organ in dogs and monkeys. In the 1-month dog study, daclatasvir induced moderate or marked decreases in the erythroid and granulocyte components of the rib and sternum in 4 pre-terminal dogs with correlating decreases in circulating leukocytes. In 2 surviving high-dose dogs, less severe (minimal or mild) marrow hypocellularity was observed. No bone marrow effects were noted at the intermediate dose (1.7-fold clinical exposure based on AUC). In the 4–month monkey study, bone marrow changes were characterised as minimal lymphoid hyperplasia (germinal center development, lymphoid follicle formation) in the rib and/or sternum of 1 male at 50 mg/kg/day and all males at 300 mg/kg/day. There were no correlating clinical pathology changes in these monkeys. The bone marrow findings were not reproducible in a 9-month study at comparable exposures (2.7-fold clinical exposure based on AUC). An explanation for this difference in study results was not provided. The applicant states that while the incidence of lymphoid hyperplasia/lymph follicle development in the bone marrow in 4 of 4 high-dose males in the 4 month study suggested a relationship to treatment, no correlative clinical pathology was evident in these animals and there were no such findings seen in the 1- and 9 month study in monkeys with comparable AUC values. In the 1- and 9- month study, the lymphoid follicle formation, also seen in 2 control animal in the 9-month study, was attributed to biological variation. This was supported by published literature which reported the presence of lymphoid follicles in the bone marrow as a background finding in monkeys and also that lymphoid nodules in the bone marrow represent a normal finding in human. Therefore explanation provided for the difference in study results was considered adequate.

Hematological toxicity is included as an important potential risk in the RMP.

Prostate and/or testes

Prostate and/or testes were additional target organs in dogs and rats at high doses. In the 1-month dog study (11.5-fold clinical exposure based on AUC), minimal or slight seminiferous tubule degeneration in the testes was observed. In the prostate gland, slight or mild atrophy of glandular epithelium was observed in 2 pre-terminal dogs. In the 1-month rat study, daclatasvir was associated with reversible decreases in absolute prostate weight without a histological correlate at the highest dose (7.1-fold clinical exposure based on AUC). This finding was not observed in the 6-month rat study at doses ≤50 mg/kg/day (≤3.9-fold clinical exposure based on AUC). Overall, given the findings in the prostate and testes of dogs occurred only in a few dogs at an overtly toxic dose and since daclatsavir was not associated with adverse effects in monkeys or impairment of fertility in rats, the effects on prostate and testes appear to be of limited concern for humans.

Other findings

In rats, treatment with daclatasvir at ≥12.5 mg/kg/day (≥0.6-fold clinical exposure based on AUC) induced large reversible increases in water consumption and urine volumes, with secondary decreases occurring in urine specific gravity, osmolality, and blood urea nitrogen. When the rats were subjected to water deprivation, the increases in urine volumes and changes in urine specific gravity and osmolality were resolved, indicating a fully-competent renal tubular urine concentrating ability. The underlying aetiology for the increased water consumption/urine volume is not understood. However, no apparent daclatasvir-related effects in serum osmolality or renal or pituitary histopathology were observed indicating that the increased urine volume was likely due to increased water consumption. According to the Applicant, there were no changes in 24-hour urine volume and serum and urine osmolality in humans

following administration of daclatasvir at doses up to the recommended human dose for 14 days that supports a lack of similar effects of in humans.

Other daclatasvir-related findings noted in repeat-dose studies mainly included findings at overtly toxic doses in the dog. These included minimal or slight pancreatic acinar cell vacuolation, and slight or mild lymphoid depletion in the thymus and/or spleen. These changes were only observed in the pre-terminal dogs and were not observed in dogs that survived to scheduled necropsy following dose reduction to 50 mg/kg/day. An additional finding in dogs was slight or mild increased extramedullary haematopoiesis at doses ≥ 15 mg/kg/day (1.7-fold clinical exposure based on AUC). At 15 mg/kg/day, this finding was reversible. At higher doses, this finding may have been secondary to decreased production or increased turnover of cells in the bone marrow. An additional daclatasvir-related finding observed only at high doses in rats included reversible multifocal discolorations of the stomach mucosa, which correlated generally with erosions and with stress. Overall, given these other findings were present mainly in early decedent dogs or were secondary to stress in rats, the risk of comparable findings in humans at the recommended human dose appears low.

Combination repeat dose toxicity

According to the SmPC, daclatasvir is indicated for combination therapy with sofosbuvir or with Peginterferon-α and ribavirin for up to 24 weeks. A 2-week repeat-dose combination study with daclatasvir, Peginterferon-α and ribavirin did not identify toxicological or toxicokinetic interactions. All findings were previously identified when the compounds were administrated alone. However, a high preterminal mortality was observed, distributed among all treatment groups, and considered caused by technical difficulties in capsule administration of ribavirin.

In line with subsequent national HA recommendations, no non-clinical combination toxicity studies were conducted with daclatsvir and sofosbuvir. However, as both sofusbuvir and daclatasvir are early stage entities with limited clinical experience and have some overlapping target organs.

In clinical studies, there have been no effects of daclatasvir on bone marrow or liver toxicity. In combination clinical studies with daclatasvir and sofosbuvir, there are no reported Grade 3/4 liver function test abnormalities, all LTFs were grade 1 or 2. ALT and AST decreased from baseline in all treatment groups. In addition, most subjects had normal haematological laboratory values. Based on above, the risk for overlapping or synergistic toxicities between daclatasvir and sofosbuvir is considered as low.

Toxicokinetics and interspecies comparison

In toxicokinetic evaluations conducted in rats, dogs and monkeys administered daclatasvir, systemic exposures to daclatasvir, BMS-805215 and BMS-795853 were determined. Generally, the systemic exposure to daclatasvir was dose-related and AUC generally increased approximately equal to or greater than dose proportional. AUC values in males and females were generally similar although in some cases exposures values were higher in females.

At the NOELs in the pivotal toxicity studies conducted with daclatasvir in mice, rats, rabbits, dogs and monkeys AUC values achieved at the NOEL or NOAEL doses were between <1 and 19x the AUC value at the recommended human dose. The applicant states that the main DCV target organs in animals with low exposure multiples were the liver and adrenal gland. No liver and adrenal gland effects have occurred in the clinical studies conducted with DCV. Therefore the clinical relevance of the low animal-to-human exposure multiples for DCV effects in animals was considered to be low. This was considered acceptable.

Genotoxicity

Daclatasvir was tested negative in a complete package of genotoxicity studies, including test for gene mutations and chromosomal aberrations in vitro and chromosomal aberrations in vivo.

Carcinogenicity

The carcinogenic potential of daclatasvir was evaluated in a 26-week study in Tg-rasH2 transgenic mice and a 2-year carcinogenicity study in Sprague Dawley rats. There were no significant increases in neoplastic changes due to daclatasvir treatment evident in either of these studies. Therefore, daclatasvir was not carcinogenic in mice at doses ≤300 mg/kg/day (≤8.7-fold clinical exposure based on AUC) or in Sprague Dawley rats at doses ≤50 mg/kg/day (≤4.6-fold clinical exposure based on AUC).

Reproduction Toxicity

In the fertility and early embryonic development study in rats, there were no effects on male reproductive parameters and the reproductive NOEL in females was 200 mg/kg/day (10-fold clinical exposure based on AUC). In male rats, there were no effects on mating performance, but reduced prostate/seminal vesicle weights and minimally increased dysmorphic sperm were observed at 200 mg/kg/day. This dose level also produced toxicity as indicated by decreased food consumption and body weight, and gross changes in the adrenals and stomach. Therefore, the male NOEL for reproductive toxicity was 50 mg/kg/day (3.4-fold clinical exposure based on AUC).

Effects in rat and dog male reproductive organs were also noted in repeat-dose toxicity studies (see prostate and/or testes). In the 9-months monkey study, evaluation of spermatogenesis by Periodic Acid Schiff staining of testes of sexually mature animals did not reveal any abnormality. However, sexual immaturity precluded an evaluation of the spermatogenesis in 12/16 monkeys.

Embryo-foetal development studies were performed with daclatasvir in rats and rabbits. The selected dose levels have greatly exceeded the MTD as evidenced by maternal mortality seen at both the intermediate and high dose levels in both species.

In the rat pivotal study, maternal toxicity was evident at ≥200 mg/kg/day as shown by termination for welfare reasons of 1 dam in each of the intermediate and high dose groups, respectively, and adverse clinical signs, body weight losses and reduced food consumption. At the highest dose (1000 mg/kg/day), a marked embryolethality (early resorptions) with associated reductions in litter size was observed. Due to profound post-implantation loss at the highest dose, the numbers of litters and live foetuses evaluated for external, visceral and skeletal malformations and variations were only 6 and 33, respectively. Statistically increased incidences of foetal malformations and associated variations were generally clustered in the fetal brain, skull, or limbs and were noted in litters at ≥200 mg/kg/day. At the highest dose, the range and severity of the malformations are consistent with a teratogenic effect throughout the organogenesis suggesting a rather non-specific mechanism. At the intermediate dose, 11/270 foetuses were classified as having malformations. According to the Applicant, there were no effects on any maternal or foetal endpoints at 50 mg/kg/day, the proposed study NOAEL. While it is agreed that the NOAEL for maternal effects is 50 mg/kg/day, the proposed NOAEL for foetal effects is not agreed with. At the low dose, an increased incidence of litters with foetuses with any malformations was observed, 13.6% versus 0% in the control group. Malformations (external and/or visceral) were seen in 3 foetuses from 3 litters and include malrotated right hindlimbs, imperforate anus, rudimentary tail and malpositioned kidneys.

In the pivotal rabbit study, the numbers of litters evaluated were even lower than in the rat study due to excessive maternal toxicity and abortions leading to dose reduction in all treatment groups already after 3 daily doses. At the high dose (750/370 mg/kg/day), 22/22 pregnant does were either found dead or sacrificed moribund and consequently, none remained for scheduled necropsy evaluations. At the

intermediate dose level (200/99 mg/kg/day), 1 doe was sacrificed moribund and 7 does were sacrificed after abortion resulting in 17 litters remaining for evaluation. Increased embryo-foetal lethality, reduced foetal body weights, and increased incidences of foetal malformations of the ribs and variations, notably affecting the developing head and skull were observed at the intermediate dose level. According to the Applicant, there were no adverse maternal or developmental effects attributed to daclatasvir at 40/20 mg/kg/day. While it is agreed that the NOAEL for maternal effects is 40/20 mg/kg/day, the proposed NOAEL for foetal effects is not agreed with. At the low dose, the incidence of litters with foetuses with any malformations was 16% versus 8.7% in the control group. This incidence was similar to that of the intermediate dose group where 17.6% foetal malformations were observed. Malformations (skeletal and/or visceral) were seen in 4 foetuses from 3 litters and include fused skull frontals, absent ribs, bent clavicles, ventricular septal defect, bulbous aorta, right-sided aorta arch, malpositioned/misshapen heart, enlarged/rudimentary right atria, malpositioned/misshapen right adrenals and testes, and malpositioned kidneys. These malformations were not observed at higher dose levels. However, based on the reduced numbers of litters evaluated at higher dose levels, a potential dose-response relationship may be masked by the profound embryolethality. In addition, numbers of foetuses with variations were also increased at the low dose, 36.1% versus 26.3% in the control group.

In conclusion, based on the available data, daclatasvir is markedly embryotoxic in rats and is considered teratogenic in both rats and rabbits. The exposure at the lowest dose level represents 4.6- and 16-fold clinical exposure based on AUC in rats and rabbits, respectively. The findings in the rat and rabbit embryofoetal development toxicity studies, including malformations in both species at the lowest dose levels tested raised concerns for use in pregnancy and in women of child-bearing potential.

The overall conclusion of the rat and rabbit EFD studies is that daclatasvir is embryotoxic and teratogenic in rats and in rabbits. The routine risk minimization measures as proposed by the applicant in the SmPC and PL are considered sufficient. The risk is included as an important potential risk in the RMP.

The prenatal and postnatal development study in rats was performed in compliance with the agreed paediatric investigation plan. Maternal toxicity was evident at the highest dose and included mortality of 1 dam during parturition, reduced body weight gains, reductions in food consumption and gross findings in adrenal glands. This dose associated with reductions in offspring birth weight and viability. At 50 mg/kg/day there were no adverse effects noted in the dams or in the F1 generation during the pre- and post-weaning period, and this dose level represents the NOAEL (2.6-fold clinical exposure based on maternal AUC).

The juvenile toxicity study was performed in compliance with the agreed paediatric investigation plan. Daclatasvir will initially be indicated for use in combination with other agents in adult patients only. Daclatasvir was clinically well tolerated by juvenile rats at oral doses ≤100 mg/kg/day (combined-sex AUC 117.9 µg·h/mL) for 10 weeks. The toxicologic profile of daclatasvir in juvenile rats was similar to that observed previously in adult rats. All daclatasvir-related changes noted at the end-of-dosing period were fully reversible after 1 month of recovery, except for adrenal vacuolation which remained unchanged in one male. Based on the lack of adrenal hypertrophy/enlargement, the NOAEL for juvenile rats was considered to be 50 mg/kg/day (3.1-fold clinical exposure based on AUC).

Toxicokinetic data

Immunotoxicity

No independent immunotoxicity studies were conducted, however immunotoxicity end-points (i.e., cytokine profiling of serum and/or liver, bone-marrow phenotyping, serum cytokine or inflammatory mediators, immunohistochemical evaluations of liver, and/or TDAR to KLH) were included in the 4- and/or 9-month monkey studies. The evaluations did not identify daclatasvir-related immunotoxicity concerns apart from decreases in mean serum interleukin-8 (IL-8) levels at Week 16/17 in the 4-month monkey

study. These effects were not considered adverse or to contribute to the safety assessment of daclatasvir because the change lacked clinical or biological relevance. The applicant states that the reduction in IL-8 levels seen in the 4-month study in monkeys was not considered adverse or clinically/biologically relevant due to the overall high variability in IL-8 levels observed generally in monkeys, and the lack of any correlation with other cytokine levels or other toxicologic findings suggestive of anti-inflammatory changes. This was supported by published literature which reported high inter-animal variability in background levels of IL-8. This was considered acceptable.

Local Tolerance

Daclatasvir was evaluated for eye (in vitro bovine cornea) and skin (rabbits) irritation, and for skin sensitization (local lymph node assay in mice) potentials. Under the conditions of these tests, daclatasvir was considered a moderate ocular irritant and a sensitizer, but was not a skin irritant.

Other toxicity studies

Phototoxicity

Daclatasvir absorbs UV light (290 to 700 nm) and bind to skin and ocular pigment of rats. In Balb/c 3T3 mouse fibroblasts in vitro, daclatasvir elicited reductions in cell viability in the presence of UVA exposure indicative of a phototoxic potential but daclatasvir was not phototoxic in pigmented rats at doses ≤100 mg/kg (7.1-fold clinical exposure based on AUC).

Dependence

No drug dependence studies were submitted. This was considered as acceptable as daclatasvir has very low distribution to the brain, no interactions were identified in secondary pharmacology screens for mechanisms associated with drug dependence and there was no evidence of effects on the central nervous system in pivotal toxicology studies.

Metabolites

No dedicated studies were conducted with daclatasvir metabolites. This was considered acceptable as there were no unique human metabolite formed in amounts above 10%.

Impurities

Potential and/or identified process impurities have been adequately assessed in Ames test in vitro and a 3-month repeat dose toxicity study in rat. The 8 investigated impurities (BMS-976332, BMS-976333, BMS-800096, BMS-800706, BMS-802783, BMS-832634, BMT-000545, and BMT-009843) are considered toxicologically qualified up to or above the proposed specification limits.

Investigative studies

In a study in dogs orally administered up to 100 mg/kg/day daclatasvir for up to 9 days, the effects seen were consistent with the previously observed bone-marrow and liver findings observed in dogs with daclatasvir. The clinical pathology changes observed identified the early onset of both bone marrow and liver lesions and support simultaneous and independent effects on both target organs following daclatasvir dosing in dogs. During the assessment, the applicant provided a discussion relating to the possible mechanisms underlying the effects seen in the liver and bone marrow from toxicity studies in monkey and dogs administered DCV. Although the investigative studies did not establish a mechanism of action for these effects the applicant states that the accumulation of DCV in hepatocytes and Kupffer cells probably caused cell proliferation and possibly inflammation in the liver. It was suggested that the material formed within the hepatic cells were probably a consequence of high levels of DCV in the liver and subsequent secretion of high levels of DCV and its metabolite into bile. The clinical relevance of this was considered to be low due to the high levels of DCV and its metabolites in dogs and monkeys dosed at 100

and 300 fold the recommended human dose (60 mg/day; 1 mg/kg/day). The bone marrow findings were observed in the dog but not monkeys treated with DCV. DCV metabolite production in dogs was found to be quantitatively higher and dissimilar to humans. The DCV metabolites were also present at higher levels in dogs than in humans or monkeys. Dogs have a different metabolite profile to humans. The bone marrow effects seen in monkeys were attributed to biological variation or spontaneous effects. In addition, no DCV-related liver or bone marrow effects were observed in the clinical studies conducted with DCV.

The discussion provided on the possible effects of DCV on the liver and bone marrow in dogs and monkeys was considered adequate by the CHMP.

2.3.5. Ecotoxicity / environmental risk assessment

A complete environmental risk assessment in accordance with EMA/CHMP/SWP/4447/00 corr 1* was submitted. Daclatasvir is considered as a persistent compound based on the long degradation half-life, however bioaccumulation was not observed in fish and therefore daclatasvir is not a PBT substance. Daclatasvir shifted significantly to sediment and a sediment toxicity study was conducted in Phase II Tier B. Daclatasvir was found to be of low toxicity to aquatic species, microorganisms and sediment dwelling organisms. It can be concluded that use of daclatasvir as indicated in the SmPC is not expected to pose a risk to the environment.

Table 1. Summary of main study results

Substance (INN / Invented Name): daclatasvir			
CAS-number (if available): 1009119-65-6			
PBT screening		Result	**Conclusion**
Bioaccumulation potential- log K_{ow}	OECD107/OECD 123	3.28 (pH 4) 4.67 (pH 7) 4.37 (pH 9)	Potential PBT (Y)
PBT-assessment			
Parameter	**Result relevant for conclusion**		**Conclusion**
Bioaccumulation	log K_{ow}	4.67	B
	BCF	6.16 – 7.05	Not B
Persistence	DT50 of ready biodegradability	Not readily biodegradable Sediment DT_{50} =187-193 days	P
Toxicity	NOEC or CMR	CMR	T
PBT-statement:	The compound is not considered as PBT nor vPvB		
Phase I			
Calculation	**Value**	**Unit**	**Conclusion**
$PEC_{surfacewater}$, default or refined (e.g. prevalence, literature)	0.3	µg/L	> 0.01 threshold (Y)
Other concerns (e.g. chemical class)			(N)
Phase II Physical-chemical properties and fate			
Study type	**Test protocol**	**Results**	**Remarks**
Adsorption/Desorption	OECD 106	K_{oc} = Soil 1 (pH 5.6) = 194 831 L/kg Soil 2 (pH 6.0) = 630 582 L/kg Soil 3 (pH 8.0) = 29 468 L/kg Soil 4 (pH 4.7) = 210 569 L/kg Sludge 1 = 2 590 L/kg Sludge 2 = 1 947 L/kg	K_{oc} for sludge below the trigger for terrestrial testing.
Ready Biodegradability Test	OECD 301B	Not readily biodegradable	
Aerobic and Anaerobic Transformation in Aquatic Sediment systems	OECD 308	Sediment 1 $DT_{50,\ whole\ system}$ =193 days Sediment 2 $DT_{50,\ whole\ system}$ =187 days > 10% shifting to sediment	Anaerobic conditions not tested Shifting to sediment triggers sediment testing
Phase IIa Effect studies			

Study type	Test protocol	Endpoint	value	Unit	Remarks
Algae, Growth Inhibition Test/*Species*	OECD 201	$NOEC_{growth\ rate}$ $NOEC_{biomass}$	1.3	mg/L	*Pseudokirchneriella subcapitata*
Daphnia sp. Reproduction Test	OECD 211	NOEC	2.3	mg/L	*Daphnia magna*
Fish, Early Life Stage Toxicity Test/*Species*	OECD 210	NOEC	0.72	mg/L	Fathead minnow (*Pimephales promelas)*
Activated Sludge, Respiration Inhibition Test	OECD 209	EC_{50}= 5 mg/L used for PNEC calculation as worst case	> 524	mg/L	
Phase IIb Studies					
Bioaccumulation	OECD 305	BCF	At 5.02 µg/L = 6.16 At 45.85 µg/L = 7.05	L/kg	Bluegill Sunfish
Sediment dwelling organism	OECD 218	NOEC	100 mg/kg	mg/kg	*Chironomus riparius*

2.3.6. Discussion on non-clinical aspects

The non-clinical development programme for daclatasvir consisted of a range of pharmacodynamic (PD), pharmacokinetic (PK) and toxicology studies, in which the activity of daclatasvir and its metabolites was investigated in vitro and in vivo. Pharmacokinetic studies detailed the absorption, distribution, metabolism, and excretion profile of daclatasvir. In the toxicity studies, daclatasvir was given orally which is the intended clinical route of administration.

The safety pharmacology parameters regarding the central nervous system and respiratory system are claimed to be evaluated in single and repeat-dose toxicity studies although the study reports do not clearly describe how the effects were studied and further clarification is requested. Cardiovascular effects of daclatasvir and metabolites were adequately evaluated in vitro and in vivo.

The non-clinical PK profile of daclatasvir was studied in mice, rats, rabbits, dogs, and monkeys. Daclatasvir showed covalent binding in liver microsomes and according to the applicant, the metabolites M20 an M21 are potentially reactive. These metabolites are considered evaluated in the non-clinical situation and the risk for potential reactive metabolite-mediated liver toxicity in humans appears acceptable. A risk for potential idiosyncratic reactions is not possible to dismiss based on available data. Daclatasvir was rapidly absorbed and extensively distributed to tissues. High concentrations of daclatasvir were located in the adrenal gland, thyroid gland, eye uveal tract, spleen thymus and kidneys. Daclatasvir binds to serum proteins to a high extent (≥95.6%). Daclatasvir reversibly bound to melanin-containing tissues. However, further examination revealed no phototoxicity concerns.

A comprehensive number of toxicology studies have been conducted to support the safety assessment of daclatasvir. The Sprague-Dawley rat and Beagle dog, which was replaced by the Cynomolgus monkey, were selected as the appropriate rodent and non-rodent species. Liver, adrenal gland and bone marrow were identified as target organs of toxicity. To date, no clinically relevant effects on liver, adrenal gland or bone marrow have been observed in clinical studies. Hepatotoxicity and haematological toxicity are included in the RMP as important potential risks.

Daclatasvir was not genotoxic or carcinogenic. Daclatasvir had no effect on fertility in rats. In embryo-foetal developmental studies, maternal toxicity (mortality, adverse clinical signs), embryolethality, reduced foetal weights, foetal malformations and variations were observed and daclatasvir is considered embryotoxic and teratogenic in both rats and rabbits. The routine risk minimization measures as proposed by the applicant in the SmPC and PL are considered sufficient. In a pre and postnatal development study, maternal toxicity and reduced F1 offspring viability were observed.

In local tolerance studies, daclatasvir was considered a moderate ocular irritant and a skin sensitizer, but not a skin irritant. No concerns are raised in terms of potential immunotoxicity or dependence potential.

The impurity profiles for the drug substance and drug product have been adequately assessed and are considered qualified up to the proposed specification limits.

2.3.7. Conclusion on the non-clinical aspects

The review of non-clinical data available for daclatasvir indicates no major issues for concern.

2.4. Clinical aspects

Introduction

GCP

The Clinical trials were performed in accordance with GCP as claimed by the applicant. The applicant has provided a statement to the effect that clinical trials conducted outside the community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

Tabular Overview of Clinical Pharmacology and Phase 1 Studies

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1/ US	AI444001	Safety, tolerability, effect on ECG and BP, PK	Randomized, double-blind, placebo-controlled, sequential, single ascending dose	Single ascending dose DCV: 0, 1, 10, 25, 50, 100, 200 mg	48	Healthy subjects	Completed
Phase 1/ US	AI444002	Safety, tolerability, PK, PD, effect on ECG and BP	Randomized, double-blind, placebo-controlled, sequential, single ascending dose	Single ascending dose DCV: 0, 1, 10, 100 mg	18	Chronic HCV GT-1	Completed
Phase 1/ US	AI444003	Safety, tolerability, effect on ECG, BP and fluid homeostasis, PK	Randomized, double-blind, placebo-controlled, sequential, multiple ascending dose	Multiple ascending dose (14 days) DCV: 0, 1, 10, 30, 60 mg QD	33	Healthy subjects	Completed
Phase 2a/ US	AI444004	PD, exposure-response, safety, tolerability, effect on BP and ECG, PK	Randomized, double-blind, placebo-controlled, sequential, multiple ascending dose	Multiple ascending dose (14 days) DCV: 0, 1, 10, 30, 60, 100 mg QD; 30 mg BID	30	Chronic HCV GT-1	Completed
Phase 1/ US	AI444005	DDI, PK, safety, tolerability	Non-randomized, open-label, single-sequence	Single Sequence DCV (2 days): 10 mg QD Ketoconazole (9 days): 400 mg QD	14	Healthy subjects	Completed
Phase 1/ US	AI444006	PK, ADME, safety	Non-randomized, open-label, single dose	Single Dose [^{14}C]-BMS-790052: 25 mg	6	Healthy male subjects	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1/ Japan	AI444007	Safety, tolerability, effect on ECG and BP, PK	Randomized, double-blind, placebo-controlled, sequential, single and multiple ascending dose	Single ascending dose DCV: 0, 1, 10, 50, 100, 200 mg Multiple ascending dose (14 days) DCV: 0, 1, 10, 100 mg QD	SAD: 40 MAD: [illegible]	Healthy male Japanese subjects	Completed
Phase 1/ US	AI444008	DDI, PK, safety, tolerability	Non-randomized, open-label, single-sequence	Single Sequence DCV (5 days): 60 mg QD Midazolam (2 days): [illegible] mg QD	18	Healthy subjects	Completed
Phase 1/ US	AI444009	Bioavailability, food effect, PK, effect of famotidine on PK, safety, tolerability	Randomized, open-label, 5-period, 5-treatment, crossover	Single Dose (5 occasions) DCV: 60 mg Famotidine: [illegible] mg High-fat meal Fasted	18	Healthy subjects	Completed
Phase 1/ Korea	AI444012	DDI, PK, safety, tolerability	Non-randomized, open-label, single-sequence, 1-way interaction	Single Sequence DCV (2 doses): 60 mg Rifampin (9 doses): 600 mg QD	14	Healthy subjects	Completed
Phase 1/ US	AI444013	Effect of hepatic impairment on PK, safety, tolerability, relationship between Child-Pugh classification and PK	Non-randomized, open-label, parallel group, single-dose	Single Dose DCV: 30 mg	30	Hepatically-impaired and healthy subjects	Completed
Phase 1/ Canada, US	AI444020	DDI, PK, safety, tolerability	Non-randomized, open-label, 3-cycle, single-sequence	DCV (10 days): 60 mg QD Ortho Tri-cyclen (67 days): Fixed dose combination	20	Healthy women of child-bearing potential	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1 US	AI444023	ECG QT/QTc, safety, tolerability, PK	Randomized, partially-blinded, placebo-controlled, positive-controlled, 4-period, 4-treatment crossover	DCV (single dose, 2 occasions): 0, 60, 180 mg Moxifloxacin: 400 mg	56	Healthy subjects	Completed
Phase 1/ US	AI444024	DDI, PK, safety, tolerability	Randomized, open-label, 2-sequence	DCV (2 days): 20 or 60 mg QD Omeprazole (7 doses): 40 mg	24	Healthy subjects	Completed
Phase 1/ US	AI444027	DDI, PK, safety, tolerability	Non-randomized, open-label, 2-treatment, single-sequence, multiple-dose, one-way interaction	DCV (10 days): 60 mg QD Digoxin (20 days): 0.125 mg QD	17	Healthy subjects	Completed
Phase 1/ Netherlands	AI444032	DDI, PK, safety	Non-randomized, open-label, 2-treatment, single-sequence crossover, multiple-dose, one-way interaction	DCV (4 days): 60 mg QD and (10 days): 20 mg Atazanavir (10 days): 300 mg QD Ritonavir (10 days): 100 mg QD	14	Healthy subjects	Completed
Phase 1/ Netherlands	AI444033	DDI, PK, safety, tolerability	Randomized, open-label, 3-treatment, multiple dose, 3-way crossover, 2-way interaction	14 days DCV: 60 mg QD Tenofovir: 300 mg QD	21	Healthy subjects	Completed
Phase 1/ Netherlands	AI444034	DDI, PK, safety	Non-randomized, open-label, 3-treatment, single-sequence, multiple-dose, one-way interaction	DCV (13 days): 60 mg and (5 days): 120 mg QD Efavirenz (14 days): 600 mg QD	17	Healthy subjects	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1/ US	AI444039	Bioavailability, food effect, PK, safety, tolerability	Randomized, open-label, 4-period, 4-treatment, crossover	Single Dose (4 occasions) DCV: 60 mg Light-fat meal High-fat meal Fasted	23	Healthy subjects	Completed
Phase 1/ US	AI444044	Absolute bioavailability, safety, tolerability	Non-randomized, open-label, single oral and intravenous dose	Single oral dose DCV 60 mg Single intravenous dose 100 µg [^{13}C,^{15}N]-BMS-790052	8	Healthy subjects	Completed
Phase 1/ US	AI444054	DDI, PK, safety, tolerability	Non-randomized, open-label, 3-treatment, single-sequence, one-way interaction	DCV (9 days): 60 mg QD Rosuvastatin (2 days): 10 mg QD	22	Healthy subjects	Completed
Phase 1/ US	AI444063	Effect of renal impairment on PK, safety, tolerability	Non-randomized, open-label, single-dose adaptive design	Single Dose DCV: 60 mg	24	Renally-impaired and healthy subjects	Completed (interim CSR available, final CSR in 2014)
Phase 1/ US	AI444064	DDI, PK, safety, tolerability	Non-randomized, open-label, 2-part, one-way interaction	Part 1 DCV (8 days): 60 mg QD Methadone (9 days): 40 - 120 mg QD Part 2 DCV (8 days): 60 mg QD Buprenorphine/ naloxone (9 days): 8/2 - 24/6 mg QD	28 (14 per part) planned	Non HCV-infected on methadone (Part 1) or buprenorphine/ naloxone (Part 2) maintenance therapy	Concluded (CSR available in 2014)

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1/ US	AI444065	DDI, PK, safety, tolerability	Non-randomized, open-label, single-sequence, 2-group, 2-way interaction	Group 1 DCV (8 days): 60 mg QD Cyclosporine (2 days): 400 mg QD Group 2 DCV (12 days): 60 mg QD Tacrolimus (2 days): 5 mg QD	28 (14 per group)	Healthy subjects	Completed
Phase 1/ Japan	AI444067	DDI, PK, safety	Non-randomized, open-label, 2-part	Part 1 DCV (14 days): 60 mg QD (7 days); 20 mg (7 days) MP-424 (12 days): 500 mg Q12h Part 2 DCV (14 days): 60 mg QD (7 days); 20 mg (7 days) MP-424 (12 days): 750 mg Q12h	30 (15 per part)	Healthy Japanese male subjects	Completed
Phase 1/ US	AI444084	DDI, PK, safety, tolerability	Non-randomized, open-label, single-sequence, 2-way interaction	DCV (12 days): 60 mg QD Escitalopram (14 days): 10 mg QD	15	Healthy subjects	Completed
Phase 1/ US	AI447009	DDI, PK, safety, tolerability, FENa	Randomized, open-label, multiple-dose	7 days DCV: 60 mg QD or ASV: 600 mg BID 14 days DCV: 30 mg QD + ASV: 200 mg BID	28	Healthy subjects	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Treated Subjects	Study Population	Study Status
Phase 1/ US	AI447039	DDI, PK, safety, tolerability	Non-randomized, open-label, 3-cycle, single-sequence	ASV (11 days): 100 mg BID[b] DCV (11 days): 60 mg QD Low-dose oral contraceptive: norethindrone acetate: 1 mg/ethinyl estradiol 20 µg (21 days) High-dose oral contraceptive: norethindrone acetate: 1.5 mg/ethinyl estradiol 30 µg (21 days)	40	Healthy women of child-bearing potential	Concluded (CSR available in 2014)
Phase 1/ US	HPC1005	DDI, PK, safety, tolerability	Randomized, open-label, 2-panel, 2-way crossover	Panel 1 DCV (14 days): 60 mg QD TMC435 (7 days): 150 mg QD Panel 2 TMC435 (14 days): 150 mg QD DCV (7 days): 60 mg QD	44 (Panel 1: 19; Panel 2: 25)	Healthy subjects	Completed

a Softgel capsule formulation administered at 100 mg BID as this dose is equivalent to tablet formulation at 200 mg BID
ADME = absorption, distribution, metabolism, and excretion; ASV = asunaprevir (BMS-650032); BID = twice daily; BP = blood pressure; CSR = clinical study report; DCV = daclatasvir (BMS-790052); DDI = drug-drug interaction; ECG = electrocardiogram; FENa = fractional excretion of sodium; GT = genotype; HCV = hepatitis C virus; MAD = multiple ascending dose; PD = pharmacodynamics; pegIFNα = pegylated-IFN alpha; PK = pharmacokinetics; QD = once daily; QT = thorough QT; RBV = ribavirin; SAD = single ascending dose; UK = United Kingdom; US = United States

Tabular Overview of Phase 2 and Phase 3 Studies

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 2b/ Australia; Canada; Denmark; Egypt; France; Germany; Italy; Mexico; Puerto Rico; Sweden; US	AI444010	Antiviral activity/ efficacy (SVR), safety, resistance	Randomized, double-blind, placebo-controlled, multinational	24 or 48 weeks DCV: 0, 20, 60 mg with pegIFNα-2a/RBV	395 (GT-1: 365; GT-4: 30)	Chronic HCV GT-1 and GT-4 (Treatment-naive)	Completed
Phase 2b/ Argentina; Australia; Canada; Denmark; France; Germany; Italy; Mexico; Puerto Rico; Sweden; US	AI444011	Antiviral activity/ efficacy (SVR), safety, resistance	Randomized, double-blind, placebo-controlled, multinational	24 or 48 weeks DCV: 0, 20, 60 mg with pegIFNα-2a/RBV	419	Chronic HCV GT-1 (Null or partial responders)	Completed
Phase 2a/ France; US	AI444014	Antiviral activity/ efficacy (SVR), safety, resistance	Randomized, double-blind, placebo-controlled	48 weeks DCV: 0, 3, 10, 60 mg with pegIFNα-2a/RBV	48	Chronic HCV GT-1 (Treatment-naive)	Completed
Phase 2a/ Japan	AI444021	Safety, antiviral activity/efficacy (SVR), resistance	Randomized, double-blind, placebo-controlled	24 or 48 weeks DCV: 0, 10, 60 mg with pegIFNα-2b/RBV	45	Japanese subjects with Chronic HCV GT-1	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 2a/ Japan	AI444022	Safety, antiviral activity/efficacy (SVR), resistance	Randomized, double-blind, placebo-controlled	24 or 48 weeks DCV: 0, 10, 60 mg with pegIFNα-2a/RBV	42	Japanese subjects with Chronic HCV GT-1	Completed
Phase 2b/ Argentina, Australia, Austria, Canada, Denmark, France, Germany, Ireland, Korea, Mexico, Netherlands, Poland, Spain, Sweden, Taiwan, UK, US	AI444026	Efficacy (SVR)/ antiviral activity, safety, resistance	Non-randomized, open-label retreatment	Prior nonresponders to pegIFNα-2a/RBV GT 1 and 4 24 weeks ASV: 200 mg (or 100 mg[a]) BID DCV: 60 mg QD with pegIFNα-2a/RBV Prior nonresponders to pegIFNα-2a/RBV GT 2 and 3 24 weeks DCV: 60 mg QD with pegIFNα-2a/RBV Treatment-naive GT 1b 24 weeks ASV: 100 mg BID[a] DCV: 60 mg QD If Rescue, up to 48 weeks	300 planned	Chronic HCV GT-1, 2, 3, and 4 Subjects must have participated in any ASV, DCV, or BMS-791325 trials and must have been assigned to the control arm with pegIFNα/RBV or placebo	Ongoing
Phase 2b/ Australia, Canada, Denmark, France, Italy, US	AI444031	Efficacy (SVR)/ antiviral activity, safety, resistance	Randomized, double-blinded, placebo-controlled	12, 16, or 24 weeks DCV: 0, 60 mg with pegIFNα-2a/RBV	151	Chronic HCV GT-2 and GT-3 (Treatment-naive)	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 3/ US, Puerto Rico	AI444038	Efficacy (SVR)/ antiviral activity, safety, PD	Non-randomized, open-label, single treatment group	24 weeks DCV: 60 mg with pegIFNα-2a/RBV ± 24 weeks pegIFNα-2a/RBV	230 planned	African American, Hispanic/Latin, White Caucasian subjects with Chronic HCV GT-1 (Treatment-naive)	Ongoing
Phase 2a/ US	AI444040	Efficacy (SVR)/ antiviral activity, safety, tolerability, DDI, PK, resistance	Randomized, open-label, parallel treatment group	12 or 24 weeks DCV: 60 mg QD PSI-7977: 400 mg QD RBV: 200 mg BID	211	Chronic HCV GT-1, GT-2, and GT-3 (Treatment-naive or TVR or BOC treatment failure)	Completed
Phase 3/ France, Greece, Italy, Puerto Rico, Spain, UK, US	AI444042	Efficacy (SVR)/ antiviral activity, safety, PD	Randomized, double-blind, placebo-controlled	24 weeks DCV: 0, 60 mg pegIFNα-2a/RBV ± 24 weeks pegIFNα-2a/RBV	120 planned	Chronic HCV GT-4 (Treatment-naive)	Ongoing

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 3/ Argentina, Australia, Belgium, Brazil, Canada, France, Germany, Italy, Russia, Puerto Rico, Spain, UK, US	AI444043	Efficacy (SVR)/ antiviral activity, safety, PD	Non-randomized, open-label, single treatment group	24 weeks DCV: 60 mg pegIFNα-2a/RBV ± 24 weeks pegIFNα-2a/RBV	300 planned	Coinfected HIV and treatment-naive chronic HCV GT-1	Ongoing
Phase 3/ Argentina, Australia, Canada, Denmark, France, Germany, Ireland, Italy, Japan, Mexico, Puerto Rico, Spain, Sweden, UK, US	AI444046	Durability of efficacy (SVR), resistance, characterization of progression of liver disease	Long-term follow-up, observational	None	1000 planned	Chronic HCV previously treated with ASV and/or DCV	Ongoing

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 3/ Argentina, Australia, Austria, Canada, Denmark, France, Germany, Israel, Italy, Poland, Russia, Spain, Switzerland, UK, US	AI444052	Efficacy (SVR)/ antiviral activity, safety, PD	Randomized, open-label	24 weeks DCV: 60 mg QD peg IFNα-2a/RBV ± 24 weeks pegIFNα-2a/RBV or 12 weeks Telaprevir: 750 mg TID peg IFNα-2a/RBV + 12 or 36 weeks pegIFNα-2a/RBV	600 planned	Chronic HCV GT-1 (Treatment-Naive)	Ongoing
Phase 2/ Argentina, France, Germany, Hungary, Spain, US	AI444062	Efficacy (SVR)/ antiviral activity, safety, PD	Randomized, open-label	GT-1b 12 or 24 weeks DCV: 30 mg QD TMC435: 150 mg QD with or without RBV GT-1a 24 weeks DCV: 30 mg QD TMC435: 150 mg QD with or without RBV	168 planned (GT-1b: 147; GT-1a: 21)	Chronic HCV GT-1 (Treatment-Naive, Null Responders to prior pegIFNα/ RBV)	Ongoing

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 2a/ France, US	AI443014	Efficacy (SVR)/ antiviral activity, resistance, PK, safety, tolerability	Randomized, open-label, parallel group, multiple-dose, dose escalation	12 or 24 weeks DCV: 30 or 60 mg QD ASV: 200 mg BID BMS-791325: 75 or 150 mg BID with or without RBV If Rescue, up to 124 weeks	320 planned	Chronic HCV GT-1 and GT-4 (Treatment-naive or null responders)	Ongoing
Phase 2a/ France, US	AI447011	Antiviral activity/ efficacy (SVR),safety, PK, resistance	Randomized, open-label, parallel group, multiple-dose	24 weeks DCV: 60 mg QD ASV: 200 mg BID with or without pegIFNα-2a/RBV If Rescue, 48 - 72 weeks	122	Chronic HCV GT-1 (Null responders)	Completed
Phase 2a/ Japan	AI447017	Safety, tolerability, efficacy (SVR)/ antiviral activity, resistance	Non-randomized, open-label, 2 parallel groups, 2 parts	24 weeks ASV: 200, 600 mg BID DCV: 60 mg QD If Rescue, 72 weeks	43	Japanese subjects with Chronic HCV GT-1 (Null responder, IFN therapy ineligible-naive/ intolerant)	Completed

Study Type/Country	Study Identifier	Primary and Secondary Study Objectives	Study Design/ Type of Control	Treatment Regimen	Number of Planned/ Treated Subjects	Study Population	Study Status/ Type of Report
Phase 3/ Japan	AI447026	Efficacy (SVR)/ antiviral activity, safety, PD	Non-randomized, open-label, 2 parallel group	24 weeks ASV: 100 mg[b] BID DCV: 60 mg QD If Rescue, 48 weeks	222	Japanese subjects with Chronic HCV GT-1b (Non-responder, IFN therapy ineligible-naive/ intolerant)	Completed
Phase 3/ Argentina, Australia, Austria, Canada, France, Germany, Ireland, Israel, Italy, Korea, Netherlands, New Zealand, Poland, Russia, Spain, Taiwan, UK, US	AI447028	Efficacy (SVR)/ antiviral activity, safety, PD	Non-randomized, open-label	24 weeks ASV: 100 mg[a] BID DCV: 60 mg QD If Rescue, up to 48 weeks	425 planned	Chronic HCV GT-1b (Null / partial responders, and IFN therapy ineligible-naive/ intolerant)	Ongoing
			Randomized, placebo-controlled	up to 24 weeks ASV: 100 mg[a] BID DCV: 60 mg QD If Rescue, up to 48 weeks	300 planned	Chronic HCV GT-1b (Treatment-naive)	Ongoing

a Softgel capsule formulation administered at 100 mg BID as this dose is equivalent to tablet formulation at 200 mg BID
ASV = asunaprevir (BMS-650032); BID = twice daily; BOC - boceprevir; CSR = clinical study report; DCV = daclatasvir (BMS-790052); ECG = electrocardiogram; GT = genotype; HCV = hepatitis C virus; IFN = interferon; MAD = multiple ascending dose; PD = pharmacodynamics; pegIFNα = pegylated-IFN alpha; pegIFNλ = pegylated-IFN lambda; PK = pharmacokinetics; QD = once daily; QT = thorough QT; qw = weekly; RBV = ribavirin; SVR = sustained virologic response; TVR - telaprevir; UK = United Kingdom; US = United States

2.4.1. Pharmacokinetics

Absorption

The absolute oral bioavailability (F) of DCV was determined to 67.0% (90% CI: 56.2 to 79.8). Based on the low hepatic extraction ratio (6%), the fraction absorbed is higher than 70%. When taken with a high-fat meal, the exposure to DCV is lowered by 25%. A light meal has no influence on exposure to DCV. Acid reducing agents (e.g. omeprazole, famotidine) reduces the exposure to DCV by 16% to 18%.

Distribution

DCV is highly protein bound (>99%). No concentration dependency in binding is seen. The fraction unbound increases from 0.5% in healthy to 1% in subjects with severe hepatic impairment. The volume of distribution at steady state, Vss, was determined to 47.1 L based on IV microdose data. In Caco-2 cells, DCV exhibited an efflux ratio of >24 suggesting that DCV is likely to be a substrate of an efflux transporter, most likely P-gp.

Elimination

Total recovery of a radioactive dose was 94%. Most of the administered dose (87.7%) was recovered in faeces, partly as metabolites (~30%) while 6.6% of the dose was recovered in urine. 95% of the radioactivity in faeces has been identified. There was one metabolite, BMS 805215 (M2), which constituted 15.2% of the dose recovered in faeces. However, it is barely observed in plasma; unchanged DCV constitutes >95% of circulating radioactivity. Metabolism and biliary/intestinal secretion of unchanged DCV mediated by P-gp and possible also other transporters are the major elimination pathways. DCV is metabolised mainly by CYP3A4 to several metabolites, none of which is considered to be important for the antiviral effect. The metabolites M20 an M21 are potentially reactive, electrophilic metabolites potentially responsible for the covalent binding to proteins observed in vitro for DCV.

Dose proportionality and time dependency

The overall data indicate that increase in exposure to DCV is near dose proportional. Time dependency was not seen in DCV trough concentration obtained repeatedly in patients during 24 weeks of treatment with 60 mg DCV once daily.

Variability

Low intra-subject (CV~10%) and moderate inter-subject (CV~35%) variability was observed in healthy volunteers. Higher inter subject (CV~50%) variability was seen in patients.

Pharmacokinetics in target population

The mean exposure to DCV comparing different treatment groups (60 mg DCV) in study AI444040 ranged from 9343 ng*h/mL to 15090 ng*h/mL. This is comparable to exposure in healthy volunteers.

Special populations

Renal impairment

The pharmacokinetics of daclatasvir following a single 60 mg oral dose were studied in non-HCV infected subjects with mild, moderate and severe renal impairment and with end-stage renal disease requiring hemodialysis. Although increases in total concentration were higher than unbound, using regression analysis of AUC vs. creatinine clearance (CLcr), daclatasvir unbound AUC was estimated to be 18%, 39% and 51% higher for subjects CLcr values of 60, 30 and 15 ml/min, respectively, relative to subjects with normal renal function. Similarly, subjects with end-stage renal disease requiring hemodialysis had a 20%

increase in unbound AUC compared to subjects with normal renal function. Based on exposure-response analyses, these increases are likely not clinically relevant.

No dose adjustment of Daklinza is required for patients with any degree of renal impairment (as presented in the SmPC sections 4.2 and 5.2).

Hepatic impairment

The exposure to DCV (based on total concentration) was roughly 40% lower in subjects with mild, moderate or severe hepatic impairment. However, when correcting for differences in plasma protein binding, the unbound exposure in moderate and severe hepatic impaired patients was comparable to healthy controls. Unbound exposure in subjects with mild hepatic impairment was still 40% lower compared to healthy controls.

The effect of other intrinsic factors such as gender, race, age or body weight does not appear to have any large effect on DCV pharmacokinetics.

Drug-Drug interactions

A thorough investigation of the drug-drug interaction potential in vivo has been performed. DCV is metabolised by CYP3A4 as well as excreted unchanged by P-gp and possible other transporters (see section Elimination). Further, P-gp and other trnasporters may also limit the absorption of DCV. It is therefore expected that strong inhibitors (e.g. ketoconazole) and inducers (rifampin) of CYP3A4 and/or P-gp will influence the exposure to DCV to a significant extent. This has been confirmed in vivo with ketoconazole showing a 3-fold increase in DCV exposure but it is unknown whether this could be specified to Pgp inhibitors or CYP3A inhibitors. Prescribing information is provided in the SmPC with respect to strong inhibitors and moderate inducers of CYP3A4/P-gp. Simeprevir coadministration resulted in double exposure of DCV, and simeprevir exposure increased by 50%. Coadministration with pegINF/RBV does not seem to have any influence on the exposure to DCV.

Daclatasvir showed relatively modest effects on the exposure to other drugs. Daclatasvir is an inhibitor of P-gp, organic anion transporting polypeptide (OATP) 1B1 and 1B3, organic cation transporter (OCT)1 and breast cancer resistance protein (BCRP). The exposure to rosuvastatin (OATP1B1/3 and BCRP substrate) was 1.5-fold increased while Cmax and AUC of digoxin were 1.65-fold and 1.3-fold increased, respectively, when coadministered with 60 mg DCV. Exposure to midazolam, a sensitive CYP3A4 substrate, was decreased by 13% following 6 days of coadministration with 60 mg DCV. The study was too short to obtain full induction.

2.4.2. Pharmacodynamics

Mechanism of action

Daclatasvir inhibits NS5A. The HCV NS5A protein (GT-1a / -1b) consists of 447/448 amino acids (AAs) and is essential for viral replication. Identification of NS5A as the drug target was based on inhibitor binding and mapping, inhibitor-induced resistant mutations and crystal structure modelling. Results indicate that daclatasvir acts at the N-terminus of the protein.

In vitro susceptibility of different genotypes in the replicon assay

The investigation of viral in vitro susceptibility to daclatasvir was mainly performed using the H77c (GT1a), Con1 (GT1b and JFH-1 (GT2a) replicons. Other viral genotypes/subtypes were studied in hybrid replicons where the NS5A sequence was replaced as relevant. The methods, techniques and replicon vectors used for describing the genotype specific activity of daclatasvir have emerged as the general standard for drug development in the field (reviewed by Lohmann and Bartenschlager J Med Chem 2013). As there is no known enzymatic activity of NS5A, cell free (enzymatic) assays were not used.

The susceptibility of different (sub)genotypes to daclatasvir was reported as follows.

(sub)genotype	EC50
-1a	0.006 nM
-1b	0.003 nM
-2a	0.01 nM
-2a with L31M substitution	4.4 nM
-2b	0.005 nM
-2b with L31M substitution	13 nM
-3a	0.26 nM
-4a	0.002 nM
-5a	0.003-0.033 nM
-6a	0.054 nM

DCV metabolites are 1-3 orders of magnitude less active than DCV on all replicons tested.

Replicon cells with resistance to DCV were obtained by maintaining the cells in the presence of this drug for 4 to 5 weeks. These changes were introduced into the wild-type replicon background and the replicon variants were tested in transient replication assays to evaluate the impact on replication ability (fitness) and contribution to resistance. The main NS5A amino acid positions were substitutions have been associated with decreased susceptibility to daclatasvir include 28, 30, 31, and 93. The impact of resistance-associated polymorphisms on daclatasvir sensitivity was shown to be genotype-specific in in vitro studies.

Effects of NS5A Substitutions on DCV Sensitivity Across Genotypes

NS5A Substitutions	DCV EC50 Values (nM)					
	GT-1a	GT-1b	GT-2a	GT-2b	GT-3	GT-4
WT	0.006	0.003	0.01	0.005	0.26	0.002
28T	3	0.05	-	-	-	-
30E	111	0.02	-	-	335	-
30H	6.5	0.02	-	-	-	1.2
30R	5.4	0.003*	0.05	0.007	-	0.02
30K	108	0.003	0.01*	0.005*	35	-
30S	0.8	0.018	-	-	0.61	0.3
31M	1.5	0.008	4.4	13	206	0.002*
31V	15	0.1	-	-	614	0.02
93H	24	0.093	35	20	1120	0.09
93C	8.2	0.006	-	-	-	0.005
28M-30S	-	-	-	-	0.61	32
30R-31M	868	0.008	-	-	-	0.02
30H-93H	410	8	-	-	-	276
92A	0.006*	0.003*	-	-	0.005	0.002*

DCV - daclatasvir; EC50 - 50% effective concentration; GT- genotype; NS5A - nonstructural protein 5A; WT -wild-type
*Polymorphism represents the GT WT sequence

The applicant has provided estimates of the frequency of naturally occurring polymorphisms that impact daclatasvir activity, based on in-house data and public databases. Of particular interest are such substitutions that significantly decrease the susceptibility to daclatasvir. For genotype 1a, the frequency of non-wildtype at each of these positions ranged from 1-7%. In particular, the frequency of polymorphisms at positions 30 and 93 was approximately 1-2% each. In genotype 1b, the Y93H polymorphism was reported at 4-9%. In genotype 2, the L31M polymorphism was detected in 60% of sequences. In genotype 3, the frequency of Y93 polymorphisms was 2.5% and A30 polymorphisms was reported at 3%.

Based on the data above, daclatasvir is anticipated to show a relatively higher barrier to resistance in genotypes -1b and 4, and lower in genotypes 1a, -2 and -3.

The mean trough concentration of daclatasvir seen in patient samples at the recommended 60 mg dose is 220 ng/mL (approximately 300 nM). It should be noted, however, that daclatasvir is 99% protein bound. The PK/PD relationship of daclatasvir is not fully understood.

Other preclinical virology findings

There is no evidence of cross resistance between daclatasvir and drugs of other classes. As anticipated given this, additive or synergistic effects have been seen in vitro with interferon alfa, an NS3/4A inhibitor, with nucleotide and non-nucleotide NS5B inhibitors, and with combinations thereof. Daclatasvir is highly selective for hepatitis C virus.

Clinical virological methods

The COBAS TaqMan HCV Test, v2.0 For Use with the High Pure System was chosen as the assay for quantitation of HCV RNA due to its wide dynamic range, low limit of quantitation/detection of HCV RNA and its accepted use within the HCV community.

The VERSANT HCV Genotype 2.0 Assay (LiPA) is a line probe assay designed to identify HCV GT-1 to 6 in human serum or EDTA plasma samples. The use of this assay was supplemented by NS5A sequencing and phylogenetic analysis.

Secondary pharmacology

Study AI444023 was a 4-way crossover TQT study in 56 subjects (only 2 were female). Daclatasvir doses (60 mg and 180 mg, administered as multiples of 30 mg tablets in the fasting state) were compared to 400 mg moxifloxacin and to placebo. Doses up to 180 mg of daclatasvir were investigated in a thorough QT study. There is no QT related signal for daclatasvir.

2.4.3. Discussion on clinical pharmacology

DCV is rapidly absorbed (t_{max} ~2 h) and has an absolute bioavailability of 67%. Exposure is slightly reduced by a high fat meal. Acid modifiers also decrease exposure due to low solubility at higher pH. However, these effects on absorption are not clinically important.

DCV elimination seems to be both biliary eliminated (35-50%) that is partly via Pgp, and metabolism mainly via CYP3A4 (35-50%). Plasma clearance is 4.3 L/h, Volume of distribution is 47 L and the half-life 10 h to 12 h. DCV is metabolized by CYP3A4 to form several metabolites, none of which contributes to efficacy. There are signs of enterohepatic recirculation and DCV has been shown to be subject to active efflux by P-gp and possible other transporters.

Ketoconazole increases the exposure to DCV which has lead to a reduction in dose under Co-treatment with potent CYP3A4/Pgp inhibitors. Strong inducers of CYP3A4 and P-gp decrease the exposure to a substantial degree and co-treatment is contraindicated. DCV is an inhibitor of P-gp, OATP1B1/3, and BCRP but has a modest influence on the exposure to other drugs *in vivo*; digoxin and rosuvastatin exposure is slightly increased due to inhibition of transporters. DCV is also an OCT1 inhibitor at clinically relevant concentration therefore an in vivo effect cannot be ruled out. DCV does not seem to inhibit any CYP to any clinically relevant extent. DCV is a weak inducer of PXR and possible also CAR pathways. The study with midazolam, a CYP3A4 substrate, was of short duration (6 days) but suggest a weak induction that is of limited clinical relevance.

Metabolism and excretion of DCV has been characterized showing that metabolism and biliary excretion is the main elimination pathways. Biliary secretion contributes to more that 25% of the elimination and it seems that other transporters than Pgp might also be involved. The applicant committed to perform a study to investigate the involvement of OCT1 as a a post-authorisation measure.

A dose modification is suggested when daclatasvir is administered in combination with strong inhibitors of CYP3A4 and/or P-gp. Simulations do suggest a stronger inhibition for CYP3A4 inhibitors with longer half-lives.

For multiple dose studies AI444003 (healthy volunteers) and AI444004 (patients), dose proportionality at steady state was rejected. Of note, when the dose was doubled from 30 mg to 60 mg to HCV patients, mean exposure increased 3.4 fold. Further, there was only a 1.2-fold increase when the dose was increased from 60 mg to 100 mg. However, due to the small study groups the results should be interpreted with caution. Over the whole dose range, near dose proportionality was observed.

Of note, DCV was co-administered with sofosbuvir in study AI444040 making interpretation of exposure in HCV patients difficult. However, in an analysis comparing to historical controls, no overt effect of sofosbvir on DCV exposure was seen. It is fair to conclude that the exposure to DCV is not essentially different comparing healthy volunteers to HCV patients.

The result from the PopPK analysis is referenced in the proposed SmPC section 5.2 Pharmacokinetics. There are claims that age, gender and race had either limited or no influence on exposure.

The exposure to DCV (based on total concentration) was roughly 40% lower in subjects with mild, moderate or severe hepatic impairment. When correcting for differences in plasma protein binding, the unbound exposure in moderate and severe hepatic impaired patients was comparable to normal controls. Unbound exposure in subjects with mild hepatic impairment was still 40% lower compared to normal controls. This finding is not explained. The exposure was variable and no evident trend can be seen in relation to Child Pugh score. It seems that hepatic impairment does not have any clinically relevant effect on unbound exposure to DCV.

The in vitro data indicate that DCV can be an inducer. A DDI study with midazolam showed a small decrease (13%) in exposure; however the study duration (6 days) may have been too short to detect full induction. In addition, there is a TDI signal in vitro. In the efavirenz (inducer) DDI study there were some indications again that CYP3A4 (or CYP3A4 and CYP2B6) was induced. Studies with oral contraceptives did not show any evidence of enzyme induction. It can be concluded that DCV is a weak inducer of PXR and possible also CAR ie CYP3A4 and CYP2B6 enzymes.

Daclatasvir is first in class as regards its mechanism of action. NS5A is considered to play a role both in viral replication and in viral assembly. Therefore, it may be that though daclatasvir has a single viral target, it in fact has more than one mechanism of action. One may speculate whether polymorphisms in NS5A might impact the effect of daclatasvir on the different NS5A actions differently. Daclatasvir shows high selectivity for hepatitis C virus.

Daclatasvir is highly potent in vitro, with picomolar EC_{50s} against genotype 1a and -1b replicons, as well as hybrid replicons representing genotypes 4a, -5a and 6a. EC_{50} values for genotype 2a varies with different expression systems, from the low picomolar to the low nanomolar, depending on the presence or absence of viral polymorphisms impacting drug susceptibility. Susceptibility for genotype 3 in vitro is also in the picomolar range, though EC_{50s} are fivefold to 250-fold higher than seen with genotype 1.

Resistance selection has been characterised in vitro. The barrier to resistance is lower in genotype 1a than in 1b, with single mutations in genotype 1a conferring over thousandfold shifts in EC_{50}. Based on in vitro data, genotype 4 seems similar to genotype -1b in terms of the relatively low impact of single amino acid substitutions. In general, across genotypes, daclatasvir is a drug that must be described as having a low barrier to resistance.

Available data indicate that there is likely cross-resistance with other NS5A inhibitors in advanced development. There is no evidence of cross resistance with drugs of other classes; furthermore, additive

or synergistic effects have been seen in vitro with interferon alfa, with a sample NS3/4A inhibitor, with nuke and non-nuke NS5B inhibitors, and with combinations thereof.

2.4.4. Conclusions on clinical pharmacology

The Clinical Pharmacology of daclatasvir has been adequately characterized in healthy volunteers and patients with hepatitis C viral infection.

2.5. Clinical efficacy

The clinical development of daclatasvir started at a time when peginterferon+ribavirin bitherapy was still standard of care for all genotypes. Therefore, dose ranging studies were performed in combination with these drugs, and the original phase II program was designed to define the best use of daclatasvir to augment the activity of a interferon-based regimen.

Subsequently daclatasvir was studied in combination with investigational NS3/4A protease inhibitor asunaprevir. This dual combination was studied in a phase II trial in which proof of concept was obtained that sustained virological response could be reached in chronic hepatitis C without the use of an interferon (Lok et al, N Engl J Med 2012). This combination is still under development, as bitherapy against genotype 1b, and as components of a tritherapy regimen with a non-nucleoside inhibitor of the NS5B polymerase.

Daclatasvir was further evaluated in a relatively large phase IIb trial (AI444040) in combination with sofosbuvir, in a cross company collaboration. The development of this drug combination was subsequently not taken into phase III, for industrial reasons. AI444040 forms the single pivotal study of this application. Since the approval of sofosbuvir, phase 3 studies of daclatasvir/sofosbuvir have started and are ongoing (SmPC section 4.4).

Overview of the main clinical trials supporting the clinical efficacy of daclatasvir

The efficacy outcomes of the following clinical trials are discussed in this assessment report

Trial number	Trial description
AI444002	Single dose phase Ib dose ranging study of DCV monotherapy in patients with genotype 1 infection
AI444004	Multiple dose phase Ib dose ranging study of DCV short term monotherapy in patients with genotype 1 infection
AI444014	Dose ranging phase IIa study of DCV in combination with pegIFN/RBV, in patients with genotype 1 infection
AI444010	Dose ranging phase IIb study of DCV in combination with pegIFN/RBV, in treatment naïve patients with genotype 1 or -4 infection
AI444011	Dose-ranging phase IIb study of DCV in combination with pegIFN/RBV in treatment experienced patients with genotype 1 infection
AI444031	Duration-ranging phase IIb study of DCV in combination with pegIFN/RBV in treatment naïve patients with genotype 2 or -3 infection
AI444040	Pivotal study for this application. Regimen- and duration comparative study of DCV in combination with sofosbuvir +/- ribavirin in patients with genotype 1, 2 or -3 infection
AI447026	DCV in combination with investigational NS3/4A inhibitor asunaprevir in patients with genotype 1b infection
AI444042	Registrational phase 3: DCV in combination with pegIFN/RBV in patients with genotype 4 infection

DCV/SOF

Clinical data from a single pivotal, open-label, randomized, Phase 2 study (AI444040, n = 211)

DCV/pegIFN/RBV

Supportive registrational studies provide exposure data to the recommended dose of DCV 60 mg QD in combination with pegIFN/RBV in 505 subjects with HCV GT-1, -2, -3, and GT-4, including 53 subjects with cirrhosis.

Study AI444010 presents data for GT-4 subjects (N = 12) treated with DCV/pegIFN/RBV. Furthermore, the applicant states that an ongoing active-controlled study AI444042 with DCV/pegIFN/RBV treatment in HCV GT-4 subjects (N = 120, 2:1 randomization, DCV/pegIFN/RBV vs placebo/pegIFN/RBV) will be available during review of the application. The study was submitted and is discussed in following sections.

DCV/Asunaprevir (ASV – an investigational NS3/4A inhibitor)

Data from another DCV regimen (DCV/ASV) from 3 completed studies are also included in this application, but are not included in the product information. These trials provide efficacy and safety data in GT-1 IFN-ineligible or intolerant patients, in prior non-responders to IFN-based therapy, and in patients with or without cirrhosis. These 3 supportive studies provide exposure data for DCV 60 mg QD/ASV in 273 subjects with HCV GT-1b.

2.5.1. Dose response studies

Initial dose ranging studies

As is typical of direct acting antivirals that are presently approved or in advanced development, daclatasvir has been dose-ranged in monotherapy and in combination with pegIFN/RBV.

In a single dose study (AI444002) of daclatasvir in patients with genotype 1 virus the median decline in log_{10} HCV RNA from baseline to 24 hours after dosing was 2.14, 3.05, and 3.40 for subjects who received DCV 1 mg, 10 mg, and 100 mg, respectively.

In a multiple dose monotherapy study (AI444004) in genotype 1a and -1b, patients with GT1a received between 1-100 mg daily in one or two doses and showed a mean maximal decrease of 4.03 log_{10} at the 60 mg dose. Those with genotype 1b received between 1-60 mg daily, and showed a mean maximal decrease of 5.65 log_{10} at the 60 mg dose. As is characteristic of drugs with a low barrier to resistance, effects were not sustained through the course of the study, due to the selection and breakthrough of resistant variants.

A further phase IIa study (AI444014) where daclatasvir was dosed in combination with pegIFN/RBV for 48 weeks was conducted in patients with genotype 1 infection without cirrhosis. SVR rates were as follows:

3 mg + pegIFN/RBV	10 mg + pegIFN/RBV	60 mg + pegIFN/RBV	Placebo + pegIFN/RBV
41.7% (5/12)	83.3% (10/12)	83.3% (10/12)	25% (3/12)

The applicant notes that although the 10- and 60-mg dose groups had similar efficacy, exposures in the 10-mg group overlapped with exposures in the sub-therapeutic 3 mg group, suggesting that subjects receiving the 10-mg dose could have exposures resulting in a sub-therapeutic response. Furthermore, no meaningful relationships between exposure and safety events were identified. Based on this data, DCV 60 mg QD was selected as the highest dose for the subsequent studies. In addition, DCV 20 mg QD was also selected for study, to minimize exposure overlap with DCV 60 mg, which provided an acceptable alternative should dose-related toxicity be observed with the higher dose.

Further dose ranging of daclatasvir in combination with pegIFN/RBV in patients with genotype 1 infection

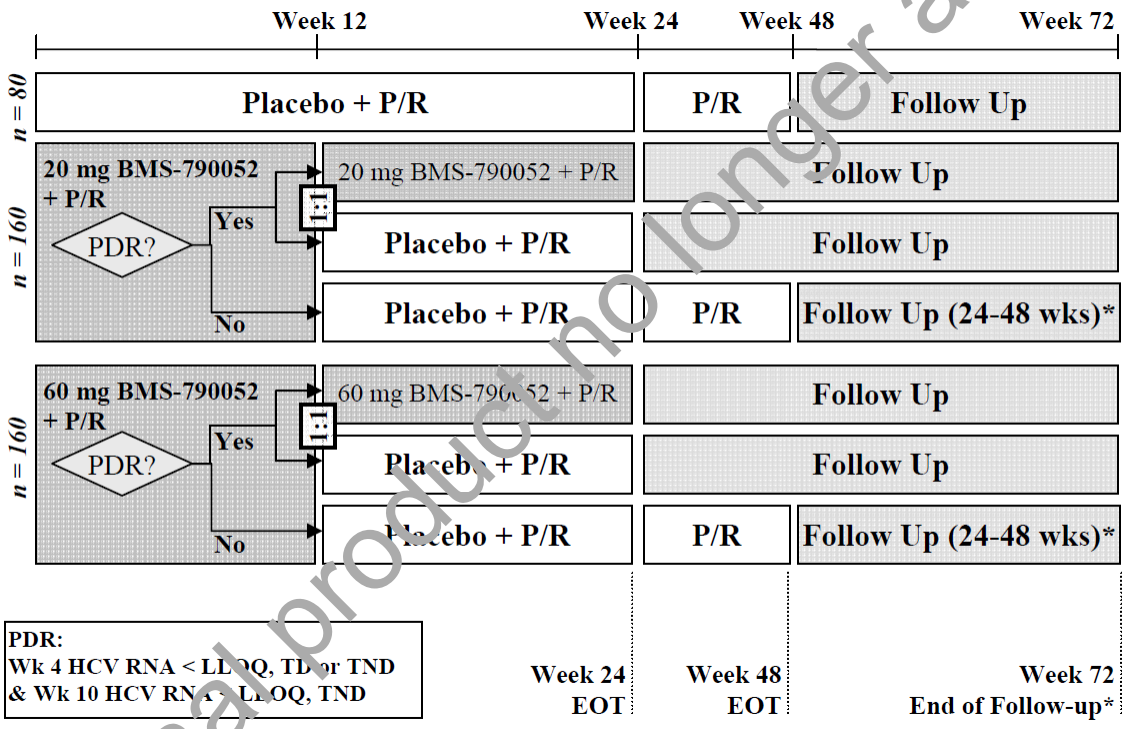
AI444010 was a randomized, double-blind, placebo-controlled, multicenter study that was conducted in treatment-naive, GT-1 and -4 HCV-infected subjects. Patients had compensated liver disease.

All subjects received DCV (20 or 60 mg)/pegIFNalfa/RBV or placebo/pegIFNalfa/RBV through Week 12.

A second randomization (1:1) occurred at Week 12 for subjects initially randomized to 20 mg or 60 mg DCV who achieved a protocol defined response (PDR: HCV RNA < LLOQ, target detected (TD) or TND at Week 4 and HCV RNA < LLOQ, TND at Week 10). These subjects either received an additional 12 weeks of DCV (20 or 60 mg)/pegIFN/RBV or 12 weeks of placebo/pegIFN/RBV.

Subjects randomized to DCV who did not achieve PDR at Week 12 received an additional 36 weeks of therapy (12 weeks placebo/pegIFN/RBV followed by 24 weeks of pegIFN/RBV) for a total of 48 weeks of therapy.

AI444010 Study Design



BMS-790052 - daclatasvir, EOT - end of treatment, P/R - peginterferon alfa plus ribavirin, PDR - protocol defined response, TD - target detected, TND - target not detected,
* Subjects assigned to 48-week DCV regimens had 24 weeks of follow-up; however, if HCV RNA was detectable at EOT or post-treatment, 48 weeks of follow-up was required

SVR24 rates in this study, with a typical design given the standards of the time, are presented by genotype, and were as follows:

Genotype	DCV 20 mg + pegIFN/RBV	DCV 60 mg + PegIFN/RBV	Placebo + pegIFN/RBV
-1a	53.8% (57/106)	54.9% (62/113)	35.7% (20/56)
-1b	73.2% (30/41)	77.4% (24/31)	43.8% (7/16)
4	66.7% (8/12)	100% (12/12)	50% (3/6)

Efficacy was considerably higher in genotype 1b compared to -1a. Virological breakthroughs were seen in 10-12% of patients with genotype 1a, compared to 2-3% in genotype 1b. Furthermore, the relapse rates

were considerably higher in GT1a (approximately 20%) compared to 1b (14%). Preclinical virological findings explain this difference, as the barrier to resistance is higher in GT1b compared to -1a. The number of patients with genotype 4 is low. However, antiviral effects of daclatasvir against genotype 4 are evident with 20/24 patients (83%) reaching SVR. Preclinical findings lead us to expect high activity in genotype 4.

There was no clear difference in the efficacy of 20 mg and 60 mg (the sample in genotype 4 being too small for conclusions).

AI444011 was a trial of daclatasvir 20 mg or 60 mg q.d., in combination with pegIFN/RBV, in genotype 1 infected patients with a history of partial or null response to pegIFN+RBV. The patients had compensated liver disease.

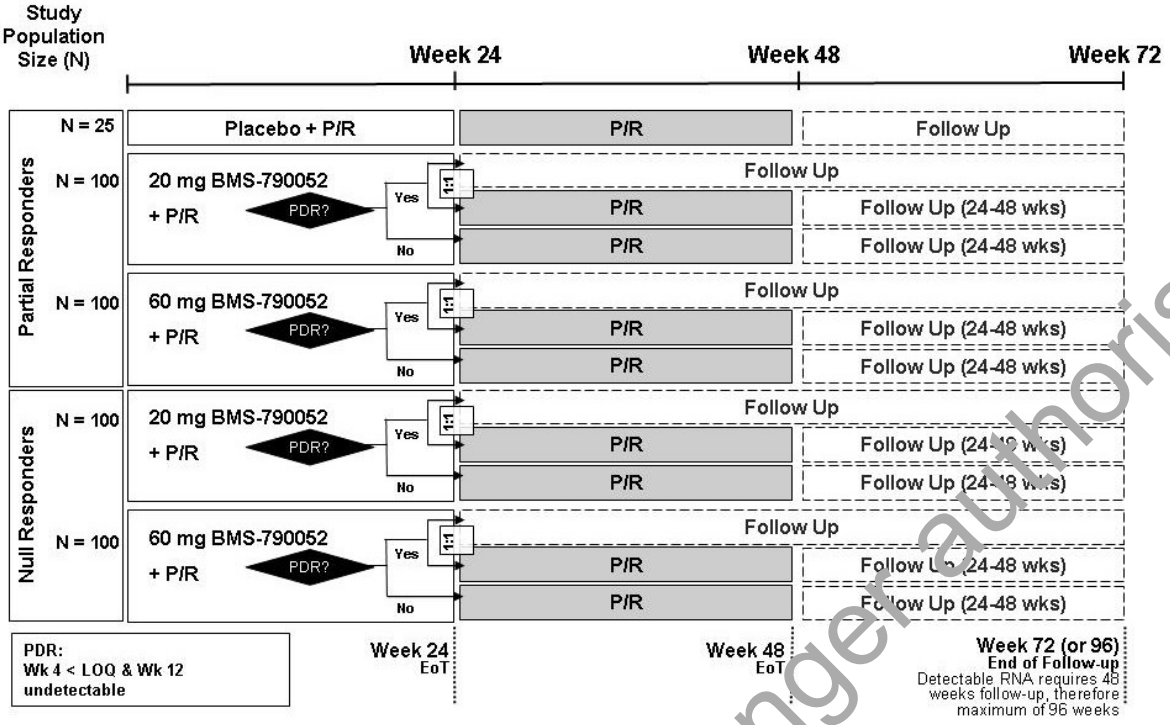
Prior null responders were randomized 1:1 to either 20-mg or 60-mg DCV QD in combination with pegIFNalfa-2a/RBV. Prior partial responders were randomized 4:4:1 to either 20-mg or 60-mg DCV or placebo QD, in combination with pegIFNalfa-2a/RBV.

A second randomization occurred at Week 24 for subjects initially assigned to 20-mg DCV/pegIFNalfa-2a/RBV or 60-mg DCV/pegIFNalfa-2a/RBV who achieved a protocol-defined response (PDR), defined as stated above, in the discussion of AI444010. Subjects who achieved a PDR were randomized (1:1) to either:

- Complete therapy at Week 24 and enter post-treatment follow up for 48 weeks (24 W DCV/pegIFNalfa/RBV group) or
- Continue therapy with pegIFNalfa-2a/RBV alone for an additional 24 weeks before entering post-treatment follow-up for 24 weeks (24 W DCV plus 24 W pegIFNalfa/RBV group)

Subjects who were randomized to DCV who did not achieve a PDR (non-PDR subjects) and subjects randomized to placebo (regardless of PDR status) received an additional 24 weeks of pegIFNalfa-2a/RBV alone for a total of 48 weeks of therapy, followed by a post-treatment follow-up period for 24 weeks.

AI444011 Study Design



Abbreviations: EoT, end of treatment; LOQ, limit of quantitation; PDR, protocol defined response; P/R, pegylated interferon alfa plus ribavirin; RNA, ribonucleic acid; Wk, week.

SVR rates were as follows:

Null responders 20 mg daclatasvir + PegIFN/RBV	Null responders 60 mg daclatasvir + pegIFN/RBV	Partial responders 20 mg daclatasvir + pegIFN/RBV	Partial responders 60 mg daclatasvir + pegIFN/RBV	Partial responders placebo + pegIFN/RBV
18.8% (25/133)	22% (29/132)	24.3% (17/70)	43.3% (29/67)	0% (0/17)

In this population with impaired interferon response, the total proportion of patients experiencing virological failure was greater with daclatasvir 20 mg q.d., compared to 60 mg q.d.. Also, as anticipated, failure rates were higher with genotype 1a compared to -1b.

As there was no difference in tolerability between 20 mg q.d and 60 mg q.d. these results supported the further investigation of 60 mg q.d. It is noted that dose ranging was only performed in genotypes 1 and 4.

Phase IIb experience of daclatasvir, in combination with pegIFN+RBV, in genotypes 2 and 3.

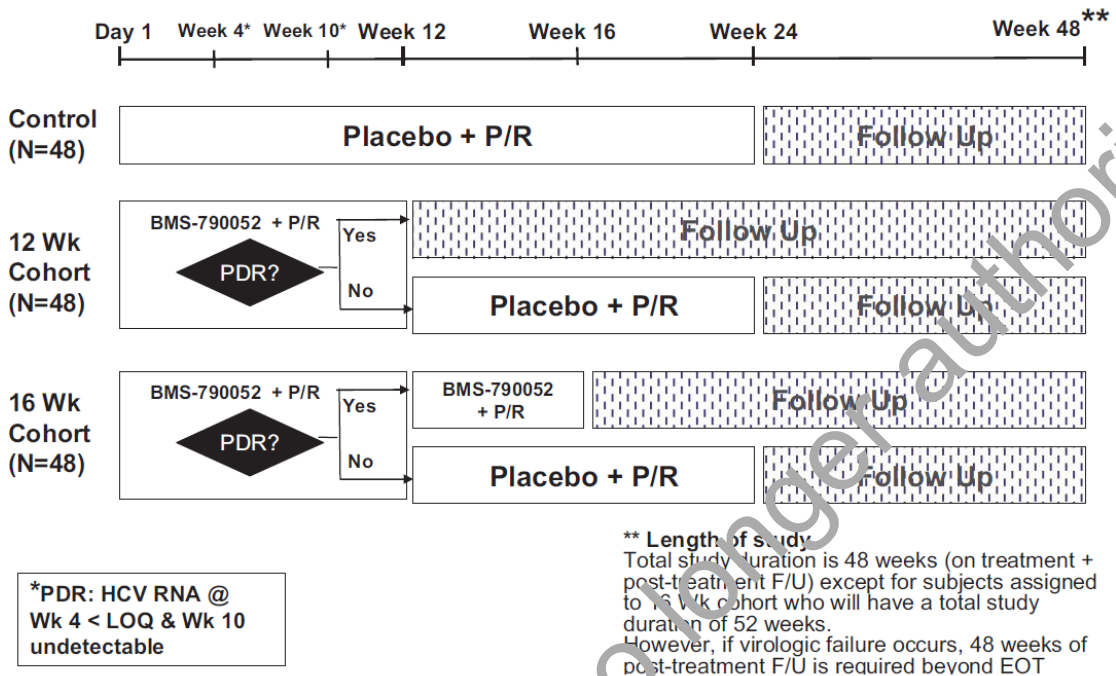
The selected dose of 60 mg daclatasvir was investigated in the AI444031 study, in treatment naïve patients with genotype 2 or -3 infection that had compensated liver disease. As SVR rates with the combination of pegIFN+RBV given for 24 weeks alone are relatively high (approximately 65-80%), the main aim of the study was to investigate whether the addition of daclatasvir might prompt a shortening of therapy.

Subjects were randomized 1:1:1 to either DCV 60 mg q.d./pegIFN/RBV for 12 weeks, DCV 60 mg q.d./pegIFN/RBV for 16 weeks, or placebo/pegIFN/RBV for 24 weeks (control group). Randomization was stratified by HCV GT determined at screening (-2 or -3). All patients received a flat dose of 800 mg RBV/day, in accordance with the ribavirin Product Information.

Subjects randomized to receive 12 or 16 weeks of DCV/pegIFN/RBV were evaluated for a PDR.

- Subjects who achieved a PDR completed 12 or 16 weeks of DCV/pegIFN/RBV therapy based on their initial randomization and proceeded to post-treatment follow-up.
- Subjects who did not achieve a PDR were required to receive 24 weeks of therapy. At Week 12 of DCV/pegIFN/RBV treatment, these subjects received an additional 12 weeks of placebo/pegIFN/RBV.

AI444031 study design



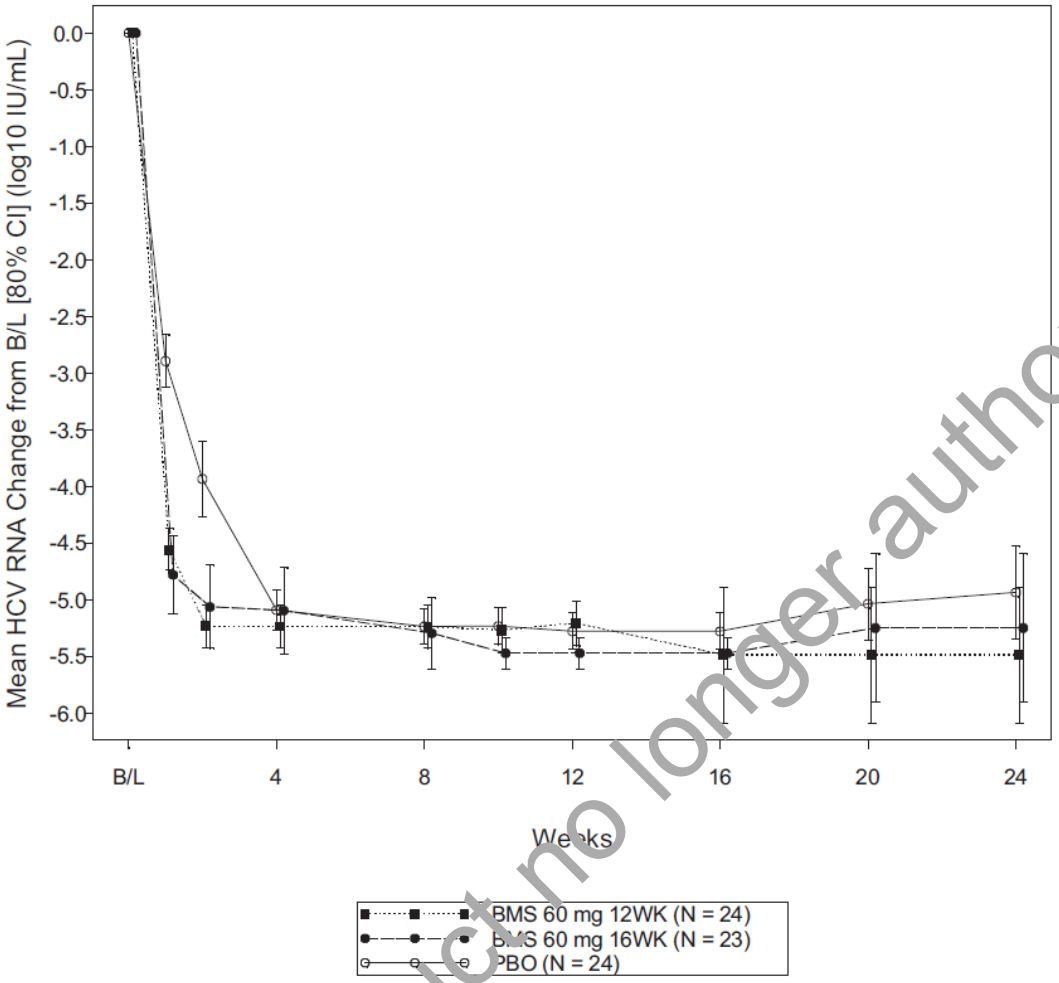
PDR is defined as HCV RNA < LLOQ, TD or TND at Week 4 and < LLOQ, TND at Week 10. In the figure, HCV RNA < LOQ is the same as HCV RNA < LLOQ, TD or TND.
BMS-790052 - daclatasvir (DCV), EOT - end of treatment, F/U - follow-up, HCV - hepatitis C virus, LLOQ – lower limit of quantitation, PDR - protocol-defined response, P/R - peginterferon alfa + ribavirin, RNA - ribonucleic acid, TD - target detected, TND - target not detected.

SVR rates in genotype 2 and -3 were as follows:

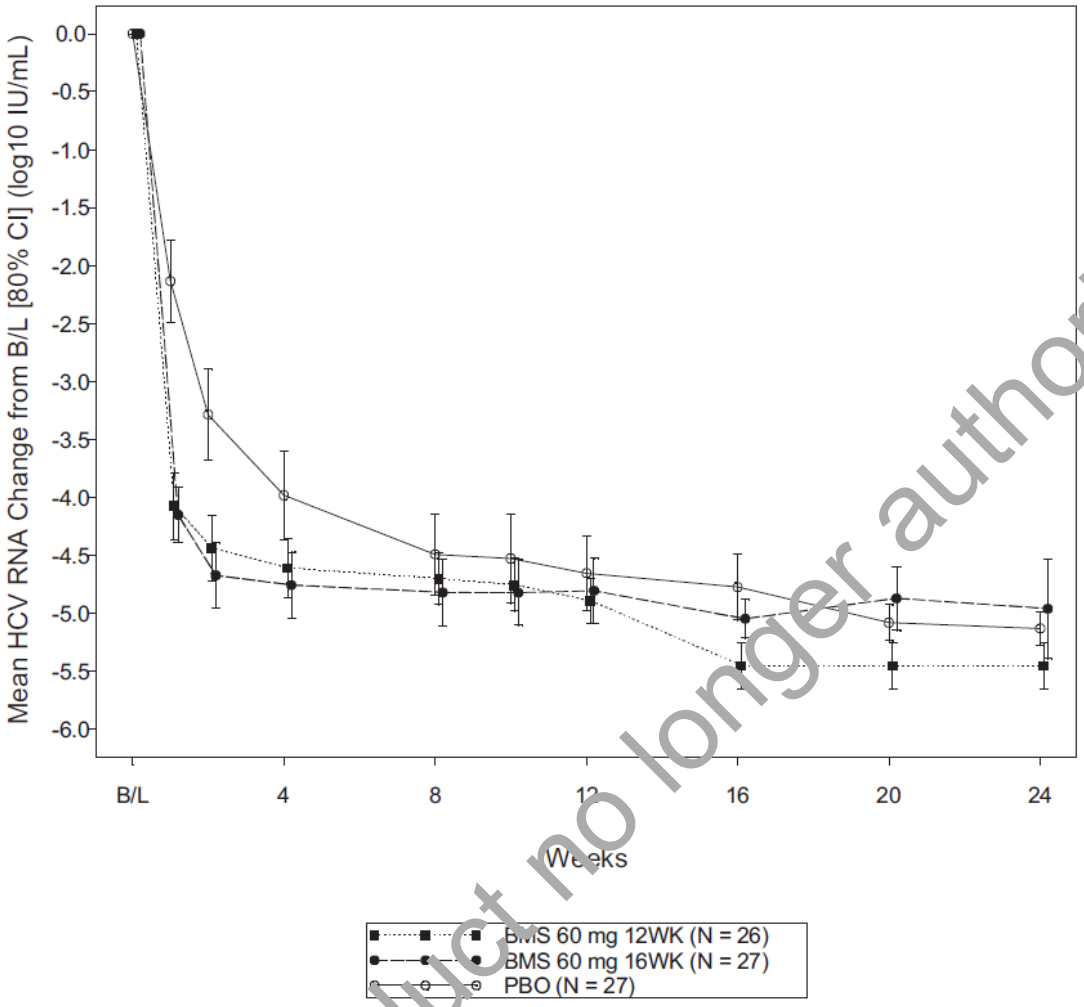
	Daclatasvir 60 mg + pegIFN/RBV, 12 weeks	Daclatasvir 60 mg + pegIFN/RBV 16 weeks	Placebo + pegIFN/RBV, 24 weeks
Genotype 2	83.3% (20/24)	82.6% (19/24)	62.5% (15/24)
Genotype 3	69.2% 18/26	66.7% (18/27)	59.3% (16/27)

The following graphs demonstrate on-treatment virological response in genotypes 2 and 3, respectively:

HCV RNA Mean Change from Baseline: HCV Genotype 2



HCV RNA Mean Change from Baseline: HCV Genotype 3



The importance of this study was that it clearly showed that daclatasvir 60 mg has antiviral activity against genotypes 2 and 3. In the light of the fact that EC_{50} values for genotype 2 are up to 6000-fold higher than those of -1b and those of genotype 3 up to 63-fold higher than those of the same reference, this would not be a foregone conclusion based on clinical studies performed in genotype 1.

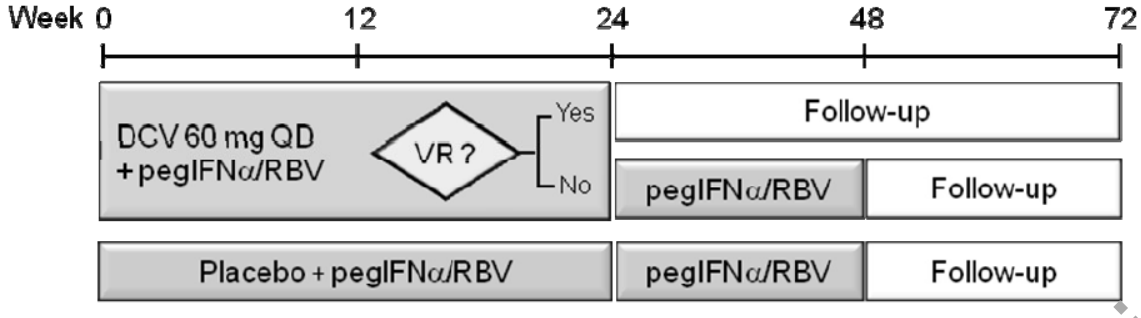
Phase 3 registrational study of daclatasvir, in combination with pegIFN+RBV, in genotype 4

AI444042 – daclatasvir in combination with pegIFN/RBV in treatment naïve patients with genotype 4 infection

During the regulatory review process, the applicant submitted results from this global multicenter study, conducted in Europe and the US. In this, adult treatment naïve patients with genotype 4 infection were randomised 2:1 to treatment with daclatasvir 60 mg once daily or placebo in combination with pegIFN/RBV. Randomisation was stratified by host IL28B C/C or non C/C genotype, and by cirrhosis status. Patients with decompensated liver disease, HIV or HBV coinfection were excluded.

Subjects treated with daclatasvir who achieved undetected plasma HCV-RNA at both week 4 and 12 completed therapy at week 24. Subjects that did not achieve such early viral response continued for an additional 24 weeks (total 48 weeks) with pegIFN/RBV. All subjects treated with placebo+pegIFN/RBV had a planned 48 week duration of therapy. The primary endpoint was SVR12.

AI444042 study design



Abbreviations: DCV - daclatasvir, pegIFNα/RBV - peginterferon alfa-2a plus ribavirin, QD - once daily, VR - undetectable (<LLOQ, TND) HCV RNA at both Weeks 4 and 12, TND - target not detected.

A total of 125 patients were randomised and 124 were treated. Study subject disposition was as follows:

Subject Disposition: All Treated Subjects

	Number (%) of Subjects		
	DCV pegIFNα/RBV	**Placebo/ pegIFNα/RBV**	**Total**
Subjects Randomized and Treated	**82**	**42**	**124**
Subjects Completing the Treatment Period	**59 (72.0)**	**26 (61.9)**	**85 (68.5)**
Reason for not Completing the Treatment Period	23 (28.0)	16 (38.1)	39 (31.5)
Adverse event	4 (4.9)	3 (7.1)	7 (5.6)
Lost to follow-up	2 (2.4)	1 (2.4)	3 (2.4)
Subject requested to discon. treatment	1 (1.2)	0	1 (0.8)
Subject no longer meets study criteria	1 (1.2)	0	1 (0.8)
Lack of efficacy	5 (6.1)	12 (28.6)	17 (13.7)
Other	2 (2.4)	0	2 (1.6)
Completed 24-wk treatment period only[a]	8 (9.8)	0	8 (6.5)
Number of Subjects Entering Follow-up	**77 (93.9)**	**40 (95.2)**	**117 (94.4)**

DCV - daclatasvir, pegIFNα/RBV - peginterferon alfa plus ribavirin

[a] Eight (8) subjects in the DCV/pegIFNα/RBV group achieved VR(4&12), completed the 24-week treatment period per protocol, and continued into the post-treatment follow-up period; however, their achievement of a VR(4&12) was not recorded in the IVRS at the Week 24 telephone call. This resulted in all 8 subjects being categorized by IVRS as not completing the study period (for the reason 'completed the 24 wk treatment period only'). Adjusting for these 8 subjects, the number of subjects in the DCV/pegIFNα/RBV group who completed the period increases from 59 to 67 (81.7%), and the number who did not complete the period decreases from 23 to 15 (18.3%).

Baseline demographic and disease characteristics were as follows:

Baseline Demographic and Disease Characteristics

	DCV/ pegIFNα/RBV (N = 82)	Placebo/ pegIFNα/RBV (N = 42)	Total (N = 124)
Age (years)			
Mean	47.7	48.4	48.0
Min, Max	20, 71	32, 61	20, 71
Age Categorization (n, %)			
< 21	1 (1.2)	0	1 (0.8)
21 - < 65 years	78 (95.1)	42 (100)	120 (96.8)
≥ 65 years	3 (3.7)	0	3 (2.4)
Gender (n, %)			
Male	61 (74.4)	29 (69.0)	90 (72.6)
Female	21 (25.6)	13 (31.0)	34 (27.4)
Race (n, %)			
White	60 (73.2)	36 (85.7)	96 (77.4)
Black/African American	18 (22.0)	5 (11.9)	23 (18.5)
Other	4 (4.9)	1 (2.4)	5 (4.0)
HCV RNA (log_{10} IU/mL)			
Mean	5.78	5.73	5.76
HCV RNA Distribution (n, %)			
< 800,000 IU/ML	43 (52.4)	26 (61.9)	69 (55.6)
≥ 800,000 IU/ML	39 (47.6)	16 (38.1)	55 (44.4)
HCV Genotype (n, %)			
1	1 (1.2)	0	1 (0.8)
4	81 (98.8)	42 (100)	123 (99.2)
Cirrhosis (n, %)			
Absent	69 (84.1)	38 (90.5)	107 (86.3)
Present	9 (11.0)	4 (9.5)	13 (10.5)
Not reported	4 (4.9)	0	4 (3.2)
IL-28B Genotype (n, %)			
CC	22 (26.8)	9 (21.4)	31 (25.0)
CT	40 (48.8)	27 (64.3)	67 (54.0)
TT	20 (24.4)	6 (14.3)	26 (21.0)
Missing			

Abbreviations: DCV - daclatasvir, HCV - hepatitis C virus, pegIFNα/RBV - peginterferon alfa plus ribavirin, RNA - ribonucleic acid

The overall high proportion of patients with plasma HCV-RNA <800,000 IU/mL is notable, but does not favour the test treatment arm.

Overall response rates, imputing SVR12 for those patients that had a later determination of SVR, was as follows, the superiority of daclatasvir over placebo being highly statistically significant:

Daclatasvir 60 mg 24 weeks + pegIFN/RBV 24-48 weeks	Placebo + pegIFN/RBV 48 weeks
81.7% (67/82)	42.9% (18/42)

The superiority of daclatasvir was consistent independent of race, region, baseline plasma HCV-RNA and IL28B genotype. In patients with cirrhosis, 7out of 9 treated with daclatasvir reached SVR, versus 1out of 4 in the placebo group.

Based on phylogenetic analyses data for DCV/pegIFN/RBV treated subjects, the SVR12 rates were high for subjects with NS5A sequences that segregated to the most common GT-4 subtypes HCV RNA GT-4a (89.3%; 25/28) and GT-4d (85.3%; 29/34); SVR12 rates were comparably high among subjects with non-GT-4a/-4d genotypes (94.1%; 16/17).

On-treatment virologic response was as follows:

HCV RNA Endpoints by Modified ITT: Treated Subjects

	Subjects with HCV RNA, Responder Evaluable (Percent)			
	DCV/pegIFNα/RBV N = 82		Placebo/pegIFNα/RBV N = 42	
Endpoint	HCV RNA < LLOQ, TD or TND	HCV RNA < LLOQ, TND	HCV RNA < LLOQ, TD or TND	HCV RNA < LLOQ, TND
Week 1	44/82 (53.7)	12/82 (14.6)	2/42 (4.8)	0/42 (0.0)
Week 2	73/82 (89.0)	37/82 (45.1)	5/42 (11.9)	4/42 (9.5)
Week 4	75/82 (91.5)	70/82 (85.4)	8/42 (19.0)	5/42 (11.9)
Week 6	69/82 (84.1)	66/82 (80.5)	17/42 (40.5)	7/42 (16.7)
Week 8	72/82 (87.8)	72/82 (87.8)	20/42 (47.6)	16/42 (38.1)
Week 12	70/82 (85.4)	69/82 (84.1)	25/42 (59.5)	20/42 (47.6)
Weeks 4 and 12 (VR 4&12)[a]	69/82 (84.1)	65/82 (79.3)	8/42 (19.0)	5/42 (11.9)
EOT	76/82 (92.7)	74/82 (90.2)	27/42 (64.3)	27/42 (64.3)
Response at follow-up Week 12	60/82 (73.2)	56/82 (68.3)	16/42 (38.1)	16/42 (38.1)

Abbreviations: DCV - daclatasvir, EOT - end-of-treatment, HCV - hepatitus C virus, ITT - intent-to-treat, LLOQ - lower limit of detection, N - number, RNA - ribonucleic acid, TD - target detected, TND - target not detected, VR - virologic response
a The Week 4 and 12 virologic response (VR[4&12]) and "Extended rapid virologic response" have the same definition, HCV RNA < LLOQ, TND at both Weeks 4 and 12

Note that the SVR12 rates in the table above do not allow for imputation of SVR12 in patients were SVR was determined later than week 12.

55/82 patients (67%) achieved an early virological response and were thus eligible for a total of 24 weeks of therapy. Among these, 94.5% achieved SVR. Among 27 patients not achieving early response, the SVR rate was 55.6%.

8/82 patients (9.8%) treated with daclatasvir experienced on-treatment virological failure, mainly categorised as virological breakthrough. The relapse rate was 2.7%.

There are some caveats in the interpretation of this study, including the high proportion of patients with a low viral load, as well as a point estimate for response in the placebo group that is lower than what is usually reported in genotype 4. Outcomes are indicative that daclatasvir has an activity against genotype 4 that is on par with that seen in genotype 1.

The appliant is proposing that a regimen of daclatasvir in combination with pegIFN+RBV could be a recommended alternative for treatment naïve- as well as –experienced patients with genotype 4 infection. Due to the side effects profile of interferon, it is generally recognised that when using such regimens, on-treatment virologic response should be monitored and treatment stopped in case of futility (to reach SVR), in order to limit non-curative exposure to interferons. The applicant has provided the following data to support stopping rules:

The majority of subjects (75/80 [94%]) in Study AI444042 had HCV RNA less than the lower limit of quantitation (< LLOQ) at Week 4 (note, 2 subjects had missing HCV RNA values at treatment Week 4 and have been removed for purposes of this analysis). The remaining 5 of the 80 subjects had HCV RNA >1000 IU/ml at treatment Week 4 and none of them achieved SVR12. At treatment week 12 no subjects had HCV RNA > LLOQ-1000 IU/ml. Three subjects had HCV RNA > LLOQ at week 12 (all 3 of these subjects had HCV RNA levels > 1000 IU/ml at week 12), none achieved SVR. This small sample forms the basis for the proposed stopping rules with this treatment modality.

2.5.2. Main study

AI444040 (pivotal trial) – daclatasvir in combination with sofosbuvir, with or without ribavirin

Title of Study

Parallel, open-label, randomized study to evaluate the safety, pharmacokinetics, and pharmacodynamics of Sofosbuvir in combination with Daclatasvir with or without ribavirin in treatment naive subjects chronically infected with hepatitis C virus genotypes 1, 2, or 3.

An addendum the trial allowed the inclusion of patients with genotype 1 virus and prior virological failure on telaprevir (TVR) or boceprevir (BOC) plus pegIFN/RBV.

Study design

This was a randomized, open-label, outpatient study with 10 treatment groups. The study was designed to be conducted in a stepwise fashion to minimize exposure of subjects to subtherapeutic treatment duration and subsequent viral resistance. Subjects were randomized separately for Groups A through F, Groups G and H, and Groups I and J. Subjects in Groups A through H were treatment-naive; subjects in Groups I and J had failed prior therapy with TVR or BOC plus pegIFN/RBV. In Groups A, C, E, G and H, and Groups B, D, and F, randomization was stratified by GT-1a and -1b and GT-2 and -3, respectively, to minimize the risk of GT imbalance between treatment regimens.

The primary objective was to estimate the rate of sustained virologic response at follow-up Week 12 (SVR12) in each treatment group, where SVR12 was defined as HCV RNA less than the lower limit of quantitation (< LLOQ, target detected (TD) or target not detected (TND) or at follow-up Week 12. This is the present standard definition of SVR in clinical trials.

Study Design for Groups A-F in Treatment-naive Subjects: 24 Weeks of Treatment

	Group	Genotype	No. of Subjects[a]	Treatment[b]		
Screening and Enrollment	A	1a/1b	15	SOF 400 mg QD x 7 days then add DCV 60 mg QD	Follow-up period (to follow subjects for 48 weeks after last dose)	Discharge
	B	2/3	16	SOF 400 mg QD x 7 days then add DCV 60 mg QD		
	C	1a/1b	14	DCV 60 mg QD + SOF 400 mg QD		
	D	2/3	14	DCV 60 mg QD + SOF 400 mg QD		
	E	1a/1b	15	DCV 60 mg QD + SOF 400 mg QD + RBV		
	F	2/3	14	DCV 60 mg QD + SOF 400 mg QD + RBV		
Days -28 to Day-1	Day 1 through Week 24				Week 24 to Week 72	Week 72 (48 weeks post-treatment)

DCV - daclatasvir, GT - genotype, QD - once daily, RBV - ribavirin, SOF - sofosbuvir (PSI-7977)
a Actual number of subjects treated
b Study drug was to be taken with a meal. Subjects meeting pre-specified criteria could have had therapeutic rescue therapy for up to 48 additional weeks (48 additional weeks for GT-1; 24 additional weeks for GT-2 and -3).

Study Design Schematic for Groups G and H in HCV GT-1 Treatment-naive Subjects: 12 Weeks of Treatment

	Group	Genotype	No. of Subjects[a]	Treatment		
Screening and Enrollment	G	1a/1b	41	DCV 60 mg QD + SOF 400 mg QD	Follow-up period (to follow subjects for 48 Weeks after last dose)	Discharge
	H	1a/1b	41	DCV 60 mg QD + SOF 400 mg QD + RBV		
Day -28 to Day-1	Day 1 through Week 12				Week 12 to Week 60	Week 60 (48 wks post-treatment)

DCV - daclatasvir, GT - genotype, HCV hepatitis C virus, QD - once daily, RBV - ribavirin, SOF - sofosbuvir (PSI-7977)
a Actual number of subjects treated
b Study drug was to be taken with a meal. Subjects meeting pre-specified criteria could have had therapeutic rescue therapy for up to 48 additional weeks.

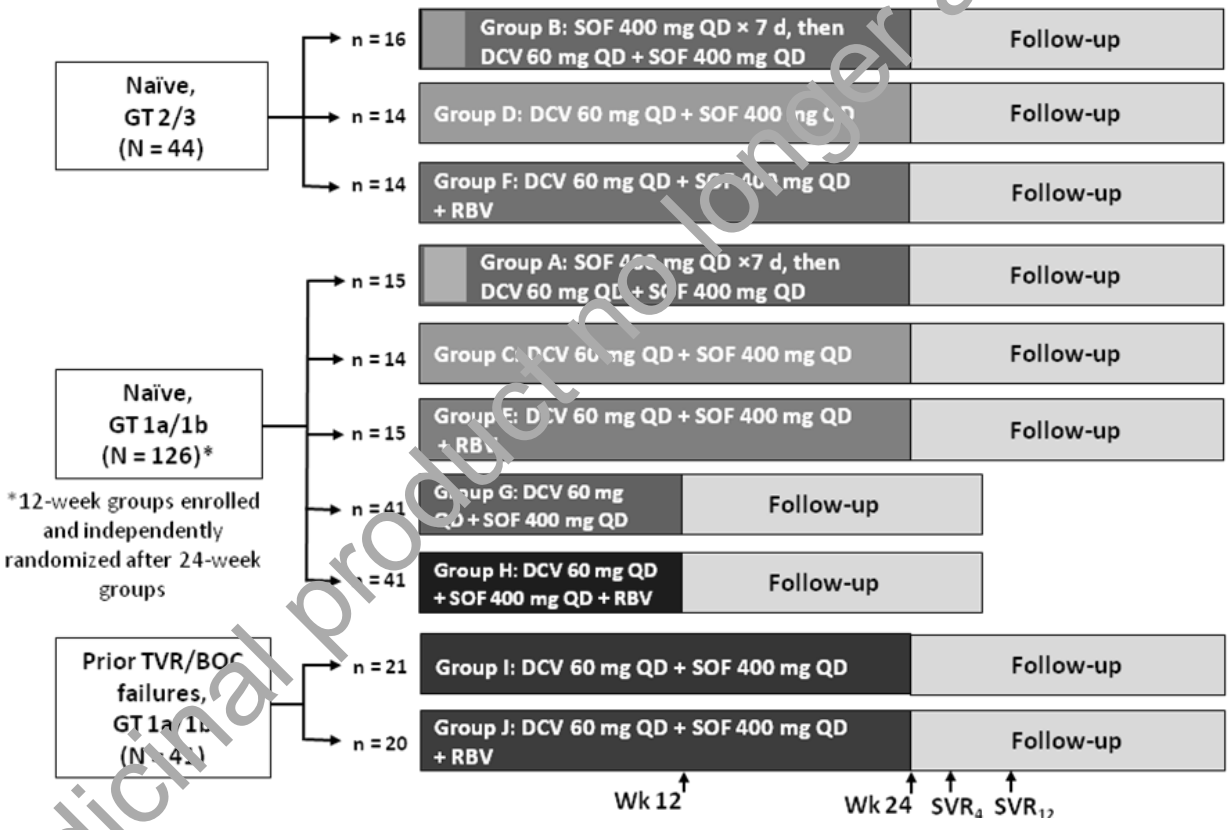
Two of the groups (A and B) had a one week lead-in with only sofosbuvir. The purpose was to build up a steady state exposure of this drug prior to daclatasvir exposure, to protect the latter from the emergence of resistant variants. No impact of this strategy was seen and the concept was dropped. Therefore, this is not further discussed and the patients in arm A and B are considered to have received a functionally similar regimen to those in arms C and D.

Study Design Schematic for Groups I and J in Subjects who Experienced TVR/BOC Treatment Failure: 24 Weeks of Treatment

	Group	Genotype	No. of Subjects[a]	Treatment[b]		
Screening and Enrollment	I	1a/1b	21	DCV 60 mg QD + SOF 400 mg QD	Follow-up Period (to follow subjects for 48 weeks after last dose)	Discharge
	J	1a/1b	20	DCV 60 mg QD + SOF 400 mg QD + RBV		
Day -28 to Day-1	Day 1 through Week 24				Week 24 to Week 72	Week 72 (48 weeks post-treatment)

As the design of this pivotal study, originally thought of as a "regimen-ranging" phase II trial, is relatively complex, the study design is also shown in the Figure below.

Study Design of Pivotal Study AI444040



This study compared treatment durations (12-versus 24 weeks) for treatment naive patients with genotype 1 infection. Furthermore, it compared treatment with daclatasvir and sofosbuvir when given as bitherapy and when given in combination with ribavirin. There was no placebo control or control with other drugs than sofosbuvir+daclatasvir. SVR in a genuine placebo control (no treatment) would have been 0.

It is important to keep in mind what might have been expected in terms of outcomes if only sofosbuvir or sofosbuvir+ribavirin had been given. The following is based on cross-study comparison.

- Sofosbuvir monotherapy is not well studied, as this treatment modality was abandoned after the phase IIa ELECTRON study in the sofosbuvir development program. The SVR rates in most genotypes would likely have been rather low with 12-24 weeks of therapy. The exception is genotype 2, where this therapeutic modality might have yielded SVR in a considerable proportion of patients.
- In genotype 1, 12 weeks of sofosbuvir+ribavirin might have yielded an SVR rate in the range of 50% in treatment-naïve non-cirrhotic subjects, such as those in the AI444040 study. When given for 24 weeks, this combination might have cured approximately 70% of patients (e.g., QUANTUM, SPARE and PHOTON-1 studies).
- In genotype 2, virtually all patients would have reached SVR with sofosbuvir+ribavirin given for 12 weeks (e.g., FISSION, POSITRON studies). Therefore, the contributory effect of daclatasvir can only be assessed in those patients that did not receive ribavirin.
- In genotype 3, sofosbuvir+ribavirin for 24 weeks might have cured up to 90% of treatment naïve, non-cirrhotic patients (e.g. VALENCE study). Therefore, also with genotype 3, it is only in those patients that received only sofosbuvir+daclatasvir that an efficacy demonstration may have been yielded in the AI444040 study.

Study Population

The study population comprised adult men and women 18 to 70 years of age with chronic HCV, a body mass index (BMI) of 18 - 35 kg/m2, inclusive, and who had the following HCV treatment history:

- Groups A through H: treatment-naive, defined as no previous exposure to an IFN formulation (i.e., IFNα, pegIFNα) or RBV; or other HCV-specific direct acting antivirals
- Groups I and J: failed treatment with a TVR or BOC containing regimen.

Subjects had HCV GT-1a, -1b, -2, or -3. Subjects co-infected with human immunodeficiency virus (HIV) or hepatitis B virus (HBV) were excluded. Subjects were to have an HCV RNA ≥100,000 IU/mL and a documented Fibrotest score ≤0.72 and aspartate aminotransferase AST): platelet ratio index (APRI) ≤2 or were without cirrhosis based on a liver biopsy within 24 months of study drug administration. Thus, patients were selected on the assumption that they were non-cirrhotic. No patients could have decompensated liver disease.

A notable consequence of the patient populations successively enrolled in this study, is that while the external validity of treatment outcomes in genotype 1 is supported by the inclusion of a demonstrably very "difficult to treat" subgroup, including patients with prior failure on telaprevir- or boceprevir triple therapy, there is no such population to inform on the validity of outcomes in genotype 2 and 3, as neither prior failures on pegIFN/RBV therapy nor cirrhotic patients were investigated.

The study was conducted in the United States, including a few sites in Puerto Rico.

Treatments

Dosing of DCV and SOF:

- All subjects were to take 2 DCV 30 mg tablets once daily (QD) and 2 SOF 200 mg tablets QD with a meal.
- For Groups A through F: A standard breakfast was to be consumed prior to dosing in the morning on Days 1 and 14 (also on Day 7 for subjects in Groups A and B) (see Table 4.3 of the protocol).

Dosing of RBV in Groups E, H, and J (subjects with GT-1):

- For subjects < 75 kg, the total dose was 1000 mg/day. Subjects were to take 400 mg (2 tablets) in the morning with a meal, and 600 mg (3 tablets) in the evening with a meal.
- For subjects ≥ 75 kg, the total dose was 1200 mg/day. Subjects were to take 600 mg (3 tablets) in the morning and in the evening with a meal. Dosing of RBV in subjects in Group F (subjects with GT-2 and -3):

For subjects infected with GT-2 and -3 the dose of RBV was 800 mg/day. Subjects were to take 400 mg (2 tablets) in the morning and in the evening with meals.

Results

Study subject disposition

Of the 211 subjects randomized and treated in Groups A through J, 207 (98.1%) completed the protocol-specified treatment period (12 weeks for Groups G and H; 24 weeks for all other groups)

2/211 patients discontinued therapy due to adverse effects. The single patients that discontinued due to "lack of efficacy" had detectable virus <LLOQ at week 8 and 10, which was subsequently not detected prior to the addition of pegIFN/RBV "rescue medication". This rescue was initiated based on very strict criteria for viral breakthrough, which were subsequently altered in a protocol amendment. There was only one patient lost to follow up.

Demographics and baseline characteristics

For treatment naïve patients with genotype 1 infection (study arms A, C, E, G, H), key baseline demographics and disease characteristics were as follows:

Age	53.5-56 (range of medians for each arm)
Gender (% male)	51% (64/126)
Race	White: 80% (100/126) Black: 17% (21/126) Other: 3% (5/126)
HCV-RNA (median)	6.09-6.79 log10 (range of medians for each arm)
Viral genotype	1a: 79% (99/126) 1b: 21% (27/126)
IL28B (C/C versus non-C/C)	C/C: 32% (40/126) Non C/C: 67% (85/126) Not reported: 1/126
Metavir class (inferred on the basis of fibrotest score	F0-F1 (minimal fibrosis): 35% (44/126) ≥ F2: 63% (79/126) Not reported 3/126

For genotype 1, prior virological failure on telaprevir- or boceprevir based therapy (treatment arms I, J) key baseline demographics and disease characteristics were as follows:

Age	57-59 (range of medians for each arm)
Gender (% male)	61% (25/41)
Race	White: 90% (37/41) Black: 7% (3/41) Other: 2% (1/41)
HCV-RNA (median)	6.31-6.35 log10 (range of medians for each arm)
Viral genotype	1a: 80% (33/41) 1b: 20% (8/41)
IL28B (C/C versus non-C/C)	C/C: 2% (1/41) Non C/C: 98% (40/41)
Metavir class (inferred on the basis of fibrotest score	F0-F1 (minimal fibrosis): 12% (5/41) ≥ F2: 83% (34/41) Not reported 2/41

For genotypes 2/3 (study arms B, D, F) key baseline demographics and disease characteristics were as follows:

Age	50-52 (range of medians for each arm)
Gender (% male)	50% (22/44)
Race	White: 86% (38/44) Black: 4% (2/44)) Other: 9% (4/44)
HCV-RNA (median)	6.73-6.92 log10 (range of medians for each arm)
Viral genotype	2: 59% (26/44) 3: 41% (18/44)
IL28B (C/C versus non-C/C)	C/C: 45% (20/44) Non C/C: 55% (24/44)
Metavir class (inferred on the basis of fibrotest score	F0-F1 (minimal fibrosis): 41% (18/44) ≥ F2: 59% (26/44)

Efficacy outcomes

Overall, the antiviral efficacy was outstanding, with >90% SVR rates in all treatment arms. This includes 40/41 patients that previously experienced virological failure with telaprevir or boceprevir in combination with pegIFN/RBV. Such patients represent demonstrably very difficult to cure patients; their inclusion and outcomes guarantee the external validity of this study in genotype 1.

All but 3 patients were <LLOQ at week 4, testifying to the potency of these regimens. No patient had genuine on-treatment virological failure; the single patient qualifying by criteria did not have quantifiable viremia. There was one established virological failure – a relapse in a patient with genotype 3 virus treated with Sofosbuvir-daclatasvir without ribavirin for 24 weeks; this patient had a baseline polymorphism which decreased susceptibility to daclatasvir.

Key HCV RNA Endpoints with DCV/SOF in AI444040 With/Without Ribavirin: All Treated Subjects

	Treatment-naive Subjects with GT-1[a]			Treatment-naive Subjects with GT-2/-3[b]			TVR/BOC Failures with GT-1[c]		
	DCV/SOF ALL N = 126	DCV/SOF With RBV N = 56	DCV/SOF Without RBV N = 70	DCV/SOF ALL N = 44	DCV/SOF With RBV N = 14	DCV/SOF Without RBV N = 30	DCV/SOF ALL N = 41	DCV/SOF With RBV N = 20	DCV/SOF Without RBV N = 21
Sustained Virologic Response (based on modified ITT analysis)									
HCV RNA < LLOQ, TD or TND									
SVR12	124 (98.4)	54 (96.4)	70 (100.0)	40 (90.9)	12 (85.7)	28 (93.3)	40 (97.6)	19 (95.0)	21 (100.0)
SVR24	120 (95.2)	53 (94.6)	67 (95.7)	41 (93.2)	13 (92.9)	28 (93.3)	41 (100.0)	20 (100.0)	21 (100.0)
SVR36	124 (98.4)	55 (98.2)	69 (98.6)	40 (90.9)	12 (85.7)	28 (93.3)	41 (100.0)	20 (100.0)	21 (100.0)
SVR48	122 (96.8)	54 (96.4)	68 (97.1)	40 (90.9)	12 (85.7)	28 (93.3)	41 (100.0)	20 (100.0)	21 (100.0)
SVR12 with imputation[d]	125 (99.2)	55 (98.2)	70 (100.0)	41 (93.2)	13 (92.9)	28 (93.3)	41 (100.0)	20 (100.0)	21 (100.0)
Relapse (cumulative through follow-up Wk 48)	1 (0.8)[e]	0	1 (1.4)[e]	1 (2.3)	0	1 (3.3)	0	0	0

BOC - boceprevir, DCV - daclatasvir, GT - genotype, HCV hepatitis C virus, ITT - intent-to-treat, LLOQ - lower limit of quantitation, RNA - ribonucleic acid, RBV - ribavirin, SOF - sofosbuvir, SVR12, 24, 36, 48 - sustained virologic response (HCV RNA < LLOQ, TD or TND) at follow-up Weeks 12, 24, 36, or 48, respectively, TD - target detected, TND - target not detected, TVR - telaprevir

a With RBV: Groups E and H; Without RBV: Groups A, C, and G

b With RBV: Group F; Without RBV: Groups B and D

c With RBV: Group J; Without RBV: Group I

d Subjects with missing HCV RNA at follow-up Week 12 were counted as SVR12 responders if they had HCV RNA < LLOQ, TD or TND at the next available measurement.

e AI444040-11-80 (GT-1a) in Group A achieved SVR4 and SVR12, then had HCV RNA 670772 IU/mL at follow-up Week 24. This subject is a likely re-infection due to viral sequences at relapse that were different from those at baseline and absence of DCV/SOF resistance detected in the virus at relapse.

Sustained Virologic Response at Follow-up Week 12 (SVR12) by Treatment Duration: Treatment-naive Subjects with GT-1

Modified ITT Analysis	DCV/SOF All	DCV/SOF With RBV	DCV/SOF Without RBV
Overall SVR12	**124/126 (98.4)**	**54/56 (96.4)**	**70/70 (100.0)**
24-week Treatment Period	44/44 (100.0)	15/15 (100.0)	29/29 (100.0)
12-week Treatment Period	80/82 (97.6)[a]	39/41 (95.1)[a]	41/41 (100.0)

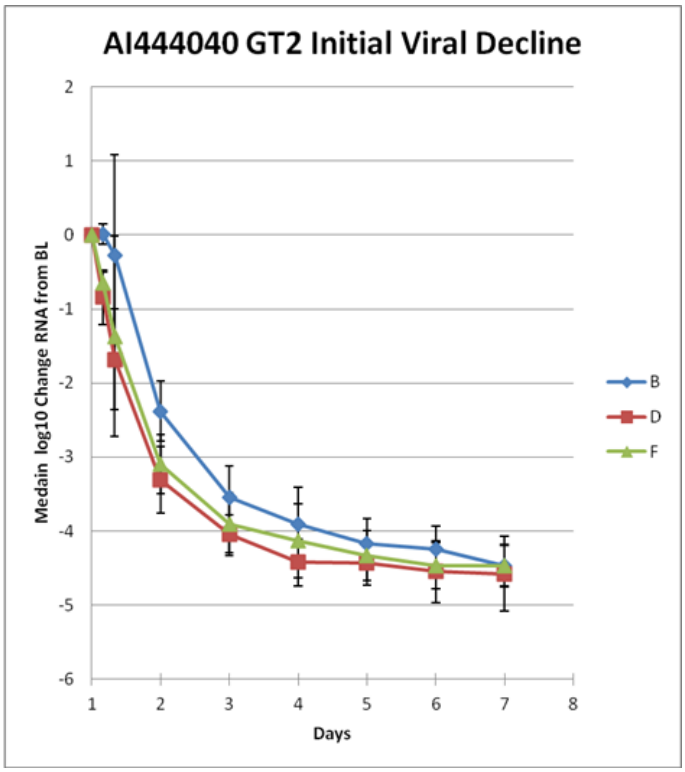
DCV - daclatasvir, GT - genotype, HCV hepatitis C virus, ITT - intent-to-treat, LLOQ - lower limit of quantitation, RNA - ribonucleic acid, RBV - ribavirin, SOF - sofosbuvir, SVR12 - sustained virologic response (HCV RNA < LLOQ, TD or TND) at follow-up Week 12, , TD - target detected, TND - target not detected
a Two subjects in Group H, who received DCV/SOF with RBV for 12 weeks, had missing HCV RNA at follow-up Week 12 and were not counted as achieving SVR12 based on the modified ITT method.

There was no apparent increase in efficacy with the addition of ribavirin. However, the size of the study precludes the firm conclusion throughout all substrata that ribavirin does not add to efficacy. Furthermore, as previously stated, the combination of sofosbuvir+ribavirin alone would have yielded a considerable effect, at least in treatment naïve patients. Also, no patients with cirrhosis were included. It is notable that all patients with genotype 2 or -3 virus were treated for 24 weeks, as were all patients with prior virological failure on NS3/4A protease inhibitor therapy.

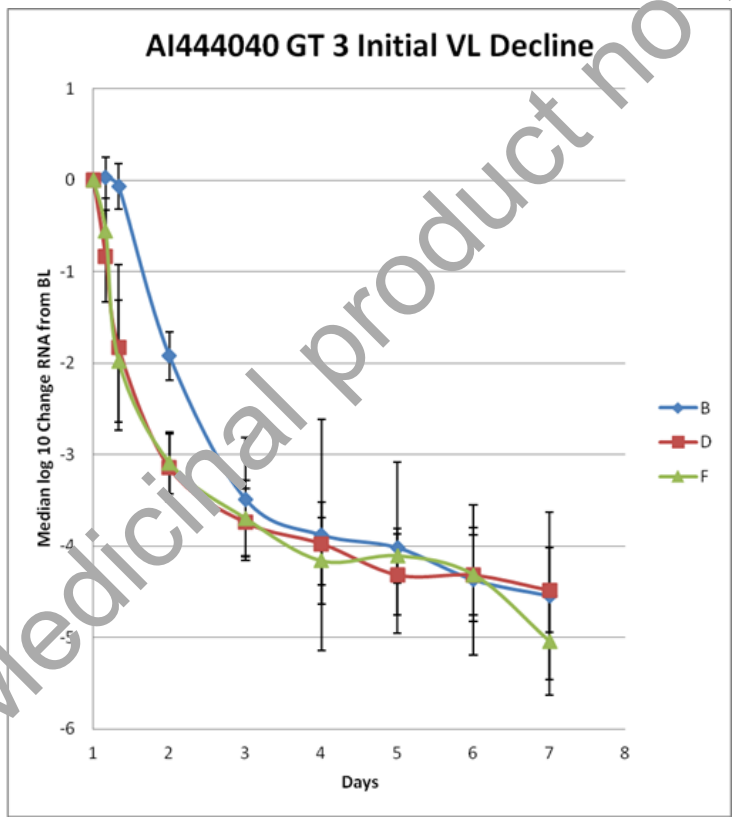
Efficacy was consistently high regardless of viral genotype or host IL28 genotype. However, the number of patients with genotypes 2 and 3 that were treated without ribavirin is very small (n=30). As patients with these genotypes treated with sofosbuvir+ribavirin for 24 weeks would likely have high response rates, it is only in the subpopulation that was treated without ribavirin that efficacy can be confidently assessed.

The contribution of daclatasvir to the efficacy of the regimen in genotypes 2 and 3 was further assessed by comparing the mean initial viral load decline in treatment groups B (where sofosbuvir was given as monotherapy the first week), D (sofosbuvir+daclatasvir) and F sofosbuvir+daclatasvir+ribavirin).

AI444040 Groups B, D F - Decline in HCV RNA in HCV Genotype-2 Subjects



AI444040 Groups B, D F - Decline in HCV RNA in HCV Genotype-3 Subjects



Along with the data previously discussed, from the AI444031 study where daclatasvir was given together with pegIFN/RBV, these graphs are indicative of the contribution of daclatasvir to the sum regimen potency in genotypes 2 and -3.

Baseline NS5A RAPs known to reduce susceptibility to inhibition by DCV in vitro were detected in 16.3% (33/203) of subjects with available NS5A sequence.

All subjects with pre-existing DCV resistance-associated variants achieved SVR, with the exception of 1 GT-3 with virologic relapse at follow-up Week 4. Resistance analysis for this subject showed an NS5A-A30K polymorphism, associated with DCV resistance, at baseline and relapse. No other resistance-associated changes were detected at relapse.

Available data are indicative of, at worst, a minor impact of common viral baseline polymorphisms on viral response. This differs from the findings when daclatsvir is used with pegIFN/RBV and are reflective of the great potency and barrier to resistance of sofosbuvir, which apparently needs relatively little support in order to achieve near maximal efficacy.

It is quite notable that the study contained seven patients with genotypes 1 or 3 virus that had estimated EC_{50s} for daclatsvir between 209-1778 nM. This represents a fold-change compared to a reference wild-type 1b replicon of 70,000 and upward. All these six patients achieved SVR.

Baseline Characteristics of Subjects with Estimated Daclatasvir EC_{50} Greater Than 200 nM who Achieved Sustained Virologic Response

Subject PID Age/Gender/ Race	Trt Gp[a]	HCV GT	Viral Outcome	NS5A RAPs at Baseline	NS5A RAPs Tested	EC50 (nM)	HCV RNA at BL (log10 IU/mL)	IL-28B GT	Fibro test Score	Metavir Score	Platelet Count at BL ($x10^9$ c/L)	VL at Day 1 (24h)	VL at Day 4	VL at WK 1	WK 2 DCV Cmin (nM)	WK 2 DCV Cmax (nM)
SOF Lead-in (DCV/SOF without RBV Treatment)																
AI444040-6-73 59/M/C	B	3a	SVR48	Y93H	Y93H	1120	7.9	C/C	0.77	F4	133	-1.94	-4.20	-4.26	234	1962
DCV/SOF without RBV Treatment																
AI444040-14-126 60/M/C	C	1a	SVR48	Q30E, Y93N	Q30E, Y93N	1778	7	C/C	0.57	F2	203	-3.32	-4.20	-4.73	89	949
AI444040-9-336 70/F/C	I	1a	SVR12	Q30[illegible], Y93H	Q30H, Y93H	589	6.3	C/T	0.46	F1-F2	152	-2.70	-3.89	-4.64	NA	NA
AI444040-8-47 36/F/P	D	3b	SVR48	A30K, L31M, S24T, S52D/E	A30K, L31M	3640	7	C/C	0.32	F1-F2	200	-2.68	-4.01	-4.48	208	2206
AI444040-18-76 55/F/C	D	3a	SVR48	S62I/T, Y93Y/H	S62I, Y93H	1412	6.6	C/C	0.57	F2	209	-2.93	-3.95	-4.69	43	1073
DCV/SOF with RBV Treatment																
AI444040-19-134 55/M/C	E	1a	SVR48	Y93N	Y93N	209	7.1	C/C	NA	NA	198	-1.45	-3.45	-4.57	212	1462
AI444040-9-105 46/M/C	F	3a	SVR48	M28M/V, S62P, Y93Y/H	M28V, S62P, Y93H	540	7	C/C	0.44	F1-F2	189	-2.83	-3.89	-5.54	141	1209

BL - baseline; Cmin - trough concentration of DCV; Gp - group; GT - genotype; HCV - hepatitis C virus; NA - not available; NS5A - nonstructural protein 5A; PID - patient identifier; Trt - treatment; WK - Week
Error! Bookmark not defined. All subjects were treated for 24 weeks.
Race: C = White; P = Native Hawaiian/Other Pacific Islander

As anticipated, prior selection of resistance against NS3/4A inhibitors did not impact response to daclatasvir+sofosbuvir +/- ribavirin.

Supportive studies

Daclatasvir in combination with investigational NS3/4A inhibitor asunaprevir in patients with genotype 1b virus

Several studies have been performed in Japan, using a combination of daclatasvir and asunaprevir. The latter drug is not yet approved and a European MAA has not been filed.

In Japan, the great majority of infections are genotype 1b. As previously discussed, daclatasvir is intrinsically more active against this subtype, compared to 1a, due to a considerably higher barrier to resistance. The same considerations apply for asunaprevir. For this reason, the dual combination was only pursued for treatment of genotype 1b, as activity towards other genotypes would be insufficient. Furthermore, in Japan the favourable IL28 CC host genotype is largely predominant. Thus, the Japanese setting in several ways represents a clinical scenario of relatively "easy to treat" patients. While there are several smaller phase II studies performed in Japan, AI447026 is large (n=222) provides the only experience of the use of daclatasvir in interferon-free combinations in cirrhotic patients, that are available in the application dossier.

In the AI447026 study, the population is HCV GT-1b, selected as being ineligible/naive/intolerant to IFN-based therapies or non-responders (null and partial responders) to peg IFN/RBV or IFN/RBV. This likely mitigates the general tendency of a Japanese treatment-naïve population to be "easy to treat". The study included subjects with baseline cirrhosis.

Patients were treated with DCV+asunaprevir (ASV) for 24 weeks. The primary objective was SVR24. Efficacy outcomes were as follows:

Efficacy Results (on treatment endpoints HCV RNA < LLOQ, TND; follow-up endpoints HCV RNA < LLOQ, TD or TND): All Treated Subjects

	Number of Subjects (%)		
Virologic Endpoints (Responder, %) Modified ITT Analysis	Non-responder (N = 87)	IFN Ineligible-naïve/Intolerant (N = 135)	Total (N = 222)
RVR	53 (60.9)	114 (84.4)	167 (75.2)
eRVR	48 (55.2)	106 (78.5)	154 (69.4)
EOTR	76 (87.4)	129 (95.6)	205 (92.3)
SVR12	70 (80.5)	119 (88.1)	189 (85.1)
SVR24	70 (80.5)	118 (87.4)	188 (84.7)
Virologic Failure	17 (19.5)	17 (12.6)	34/222 (15.3)
Virologic Breakthrough	10 (11.5)	4 (3.0)	14/222 (6.3)
Relapse* (in subjects who were HCV RNA < LLOQ, TND at EOT)	6/76 (7.9)	11/129 (8.5)	17/205 (8.3)

Virologic breakthrough on DCV/ASV therapy was observed in 4/135 (3.0%) GT-1b subjects and virologic relapse following HCV RNA < LLOQ, TND at EOT was observed in 11/129 (8.5%) subjects that were interferon intolerant/ineligible.
Viral breakthrough on DCV/ASV therapy was observed in 10 (11.5%) GT-1b subjects and virologic relapse following HCV RNA < LLOQ, TND at EOT was observed in 6/76 (7.9%) GT-1b subjects that were prior non-responders.

The efficacy of daclatasvir in patients with cirrhosis

The pivotal AI444040 excluded patients that were deemed to have cirrhosis at baseline, based on biopsy

previously performed in clinical practice.
The number of cirrhotics in studies of daclatasvir with peginterferon and ribavirin was low, precluding a real estimation of the efficacy of this regimen in such patients. However, as previously stated, interferon based regimens are no longer pursued in the development of daclatasvir, and the clinical relevance of such findings are low, as use of this regimen is not anticipated. In the application, outcomes were reported from 22 cirrhotics treated with DCV/ASV.

SVR24 by Baseline Cirrhosis: DCV/pegIFNα/RBV Regimen

	Number (%) of Subjects			
	Cirrhotic		Non-cirrhotic	
	DCV/pegIFNα/RBV	Placebo/pegIFNα/RBV	DCV/pegIFNα/RBV	Placebo/pegIFNα/RBV
	% (number of subjects/total)			
Treatment Naive				
AI444010 (GT-1)	62.5 (5/8)	37.5 (3/8)	59.9 (82/137)	37.5 (24/64)
AI444031 (GT-3)				
12 week treatment	42.9 (3/7)	-	78.9 (15/19)	-
16 week treatment	50.0 (2/4)	-	75.0 (15/20)	-
24 week treatment	-	42.9 (3/7)		65 (13/20)
AI444011 (GT-1)				
Partial Responders	50 (7/14)	0 (0/3)	41.5 (22/53)	0 (0/14)
Null Responders	10.0 (2/20)	-	24.1 (27/112)	-

SVR24 by Baseline Cirrhosis: DCV/ASV Regimen Study AI447026 (GT-1b)

	Cirrhotic	Non-Cirrhotic
Prior Non-Responders	90.9 (10/11)	78.9 (60/76)
IFN Ineligible-Naive/Intolerant	90.9 (10/11)	87.1 (108/124)
Total	90.9 (20/22)	84 (168/200)

Clinical drug resistance

As shown above, baseline polymorphisms impacting the EC_{50} of daclatasvir are common; however, they did not appear to impact response in the AI444040 study, as discussed above.

Concerning genotype 1, a relation between baseline polymorphisms at 28, 30, 31 and 93 position and an increased rate of virological failure was apparent in patients treated with daclatasvir+pegIFN/RBV, particularly in those with prior pegIFN/RBV experience. The prevalence rates for polymorphisms at each of these sites varied between studies and subgenotypes, with particular polymorphisms seen in up to 14% of some datasets. In the AI447026 study, where daclatasvir was used in combination with asunaprevir in genotype 1b, there was an association between baseline polymorphism reducing susceptibility to daclatasvir and virological failure. In particular, Y93H was present at baseline in 14% of patients; 57% of these failed therapy; overall 20.4% of patients with baseline resistance associated mutations that were treated with daclatasvir+asunaprevir failed therapy, compared to 8.7% of those without. These data indicate that, as anticipated, resistance associated substitutions likely have a different impact depending on the potency and barrier to resistance of the co-treating agents.

In patients with genotype 2 virus participating in study AI444031 NS5A-F28C/L was detected in 68.2% (30/44) of subjects with available baseline NS5A sequence. NS5A-L31M was detected in 52.3% (23/44)

of subjects with available baseline NS5A sequence of whom 17.4% (4/23) were defined as treatment failures. There appears to be a relation between the pre-existence of the conserved polymorphisms F28C/L L31M and a higher risk of virological failure when using daclatasvir in combination with pegIFN+RBV, though the numbers are small. Of note, all virological failures in GT2 had baseline resistance mutations.

The conserved L31M polymorphism produces a 146-fold shift in EC_{50} for GT2a and a 12800-fold change in the susceptibility in GT2b.

The impact of the common polymorphisms in genotype 2, on initial viral decline when using daclatasvir in combination with pegIFN/RBV in the AI444031 study was analysed.

Median Decline in HCV RNA in Subjects Treated with DCV/PegIFNα/RBV Versus placebo/PegIFNα/RBV

1. Treatment Duration	2. Baseline NS5A Variant	3. Median WK1 Change from Baseline	4. Subjects (n)
DCV WK 12/16 Subjects	F28C/L	-5.010	18
DCV WK 12/16 Subjects	L31M	-4.845	11
DCV WK 12/16 Subjects	F28L, L31M	-4.218	11
DCV WK 12/16 Subjects	No F28C/L or Y93H RAP	-4.927	2
PBO 24 WK	Mixed	-0.985	22

Abbreviations: DCV - daclatasvir; HCV - hepatitis C virus; NS5A - nonstructural protein 5A; PegIFNα - Pegylated interferon alfa; PBO - placebo; RBV - ribavirin; STDV - standard deviation; WK - week.

These data are indicative that daclatasvir retains clinically meaningful activity in the presence of L31M in genotype 2.

In patients with genotype 3, examination of the baseline NS5A RAPs at positions 30 and 93 revealed a potential association with virologic outcome when comparing their natural prevalence. Of the 8 subjects with NS5A-A30K (EC50 or NS5A-Y93H, 50% (4/8) relapsed. As shown above, these variants incur numerically significant shifts in the EC_{50}. In the AI444040 study there was no clear difference between the initial viral load declines depending on the presence of polymorphisms at 30 and 93 positions, though it is recognised that numbers are small. When daclatasvir was used in combination with pegIFN/RBV, however, mutations at these positions decreased but did not abolish the contribution of daclatasvir to initial regimen potency.

In study AI444010, NS5A RAPs were detected in 100% (13/13) of patients with genotype 4 infection, and included L28M, L30R, M31V, H54R, P58A/T, and D62E/Q. Of the 13 subjects with baseline NS5A RAPs, no subject experienced virologic failure. In the AI444042 study NS5A RAPs at positions 28, 30, 31 or 58 were seen in 71% of patients, 73% of whom achieved SVR. Susceptibility analysis of reference GT-4 replicons harboring NS5A resistance-associated substitutions revealed DCV EC50 values ranging from 0.002 to 0.9 nM while the DCV EC_{50} value against the reference GT-4 strain was 0.002 nM. It is notable that the impact on susceptibility of baseline polymorphisms detected in GT4 is considerably smaller than in GTs 2 and 3. Virological failure with daclatasvir-containing regimens is associated with the selection of variant with reduced susceptibility to daclatasvir. In general, the resistance mutations emerging in the clinic were identified in preclinical selection experiments (positions 30, 31, 62, 93).

Long-term follow-up study

A long-term follow-up study (AI444046) is ongoing to assess the durability of virologic response up to 3 years. Interim data from this study are indicative that the durability of SVR12 reached with daclatasvir containing therapy is similar to that previously seen with other treatment modalities.

Furthermore, the persistence of resistant variants selected on treatment failure is ongoing. It is notable that over 24-48 weeks of therapy the viral population tends not to revert to baseline/wild-type. This is indicative that the resistant variants are as fit as wild-type in vivo, notwithstanding in vitro replication capacity studies that imply otherwise (data not shown). The finding that reversion is rare differ from those seen with NS3/4A inhibitors (particularly in genotype 1b) and nucleotide NS5B inhibitors, where the major population tends to revert to wild-type at a variable rate after the cessation of selection pressure.

Summary of main studies

The following tables summarise the efficacy results from the main studies supporting the present application. These summaries should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment.

Table with Summary of efficacy per trial (Please refer to Appendix 1 of this document).

2.5.3. Discussion on clinical efficacy

Design and conduct of clinical studies

The scope of the efficacy demonstration

The following studies on clinical efficacy have been discussed above, and form the basis of the evaluation of the dose-response and efficacy of daclatasvir:

Studies AI444002, AI444004 and AI444014 were performed in patients with genotype 1, where daclatasvir was given as monotherapy or in combination with pegIFN/RBV, in a range of doses from 1 mg to 100 mg to guide the dose selection.

Studies AI444010 and AI444011 were larger studies performed in patients with genotype 1 virus, as well as a few patients with genotype 4. Daclatasvir was given at doses of 20 mg or 60 mg in combination with pegIFN/RBV. On this further basis the proposed dose was selected.

Study AI444031 was performed in patients with genotype 2 or 3 virus. Patients received 60 mg daclatasvir in combination with pegIFN+RBV. Results support the clinical activity of daclatasvir against these genotypes.

Study AI444012 was performed in patients with genotype 4. Patients received 60 mg daclatasvir in combination with pegIFN+RBV. Results indicate that the efficacy of daclatasvir in genotype 4 is comparable to that in genotype 1.

Study AI444040 is pivotal to this application. It was performed in patients with genotype 1, 2 and 3 virus that were deemed not to have cirrhosis. It included two cohorts of genotype 1 patients that had previously failed telaprevir or boceprevir based triple therapy. In this study, daclatasvir 60 mg was given in combination with sofosbuvir, with or without ribavirin.

Study AI447026 was a study performed in Japanese patients with genotype 1b virus. Daclatasvir was given in combination with investigational NS3/4A inhibitor asunaprevir. It is of importance to this application as it is the only study where daclatasvir was given to cirrhotics in an interferon-free treatment combination.

Comments on the design and conduct of the development program

The daclatasvir development program spans two different treatment paradigms for hepatitis C virus infection. Originally, daclatasvir was developed for use in combination with pegIFN/RBV as part of a triple therapy regimen, or in combination with an NS3/4A inhibitor as a quadruple regimen. Within the scope of the development program, however, proof of concept that hepatitis C virus clearance could be reached without an interferon was delivered in a small study of daclatasvir+asunaprevir bitherapy (Lok et al, N Engl J Med 2012). Subsequent to that demonstration, the field of hepatitis C therapy has been radically transformed, and interferon-based regimens are anticipated to be a historical phenomenon within short

Thus, after phase I monotherapy, daclatasvir was developed through phase II in combination with pegIFN/RBV. These studies were designed according to relevant standards and largely in conformity with regulatory advice. They generally employed a peginterferon+ribavirin bitherapy comparator arm.

As the field turned interferon-free, daclatasvir was investigated in combination with the aforementioned asunaprevir, primarily in Japan (but also in global sites), as the prevailing genotype in that country is -1b, and it is only for this subtype that high SVR rates could be anticipated with this combination.

In the pivotal study of this application, daclatasvir was used in combination with nucleotide NS5B inhibitor sofosbuvir, with or without ribavirin, in patients with genotype 1, -2 or -3 infection. This study was a cross company collaboration. The interferon free studies were conducted either as non-comparative studies or as dose and regimen comparative trials. This is in agreement with advice given by the CHMP.

Efficacy data and additional analyses

Dose selection

The dose of 60 mg q.d. was selected on the basis of a number of phase I and IIa trials in patients with genotype 1 infection. It is anticipated to yield exposures compatible with maximal efficacy against this genotype; furthermore, its safety profile is apparently no different from other doses tested. Daclatasvir has not been dose-ranged in monotherapy in other genotypes. 20mg and 60mg in combination with pegIFN+RBV were compared in a small sample of patients with genotype 4 infection; the results are supportive of the 60 mg dose.

Genotype 1

In the pivotal AI444040 study, daclatasvir in combination with sofosbuvir, produced SVR12 in 164/167 patients with genotype 1, with no confirmed virological failures. The population included 41 patients with previous virological failure when using telaprevir or boceprevir in combination with pegIFN/RBV. The outstanding results in this subpopulations, where all patients were demonstrated to have reached either SVR12 or SVR24 demonstrate that the general efficacy seen in patients with genotype 1 in AI444040 is not due to the selection of “easy to treat” patients – that is, the external validity of genotype 1 outcomes.

Genotype 2

The efficacy of daclatasvir against genotype 2 has been investigated in combination with pegIFN/RBV in the phase II AI444031, and in combination with sofosbuvir +/- ribavirin in the pivotal AI444040. In genotype 2, L31M is a conserved polymorphism which confers reduced susceptilbility to daclatasvir, and which were present at baseline in 60% of patients with genotype 2. Overall, outcomes of AI444031 are indicative that daclatasvir has relevant antiviral activity against genotype 2, and also so in the presence of the L31M polymorphism.

In AI444040, patients with genotype 2 were treated for 24 weeks. Furthermore, 9/26 patients with genotype 2 were treated in combination with both sofosbuvir and ribavirin. Emerging data from the

sofosbuvir development program has shown that almost all genotype 2 patients treated with sofosbuvir and ribavirin alone for 12 weeks reach SVR. Therefore, the efficacy demonstration of daclatasvir+sofosbuvir in genotype 2 rests on a mere 17 patients treated in the ribavirin-free arms. Notably there were no virological failures among these patients. Viral kinetic data are indicative that daclatasvir is contributing to the antiviral effect of the regimen. However, it cannot be stated, based on available data, that the addition of daclatasvir to sofosbuvir+ribavirin would meaningfully increase the response rate, as this is already near 100%. Furthermore, there are presently no data to support a proposed regimen of sofosbuvir+daclatasvir for 12 weeks.

Genotype 3

Evidence on efficacy in genotype 3 is similar in extent to that in genotype 2. There are data from the phase II AI444031 trial, as well as a small sample in the AI444040 study. A total of 18 patients were treated for 24 weeks in the AI444040 study; five of these received supplementary ribavirin. As the anticipated cure rate with a sofosbuvir+ribavirin bitherapy regimen for 24 weeks is high in treatment naïve, non-cirrhotic patients, there thus remain 13 patients on which to base conclusions. As is the case with genotype 2 - and in contrast to genotype 1 - there is no evidence of efficacy in confirmed “difficult to treat” patients.

Given the scarcity of data in genotype 3, any conclusions must also rest on bridging via preclinical susceptibility viral kinetics and resistance data. The average EC50 for genotype 3a was reported as 0.25 nM. This may be compared to 0.01 nM for wildtype GT2a, 0.006 nM for GT1a and 0.003 nM for GT1b. While the value for GT3a thus is higher than that for those where a larger clinical efficacy demonstration is available, it should be recognised that daclatasvir appears to have contributed to regimen efficacy in genotype 1a for a number of patients with baseline polymorphisms conferring considerably higher EC50 values compared to that for genotype 3. Viral kinetic data are indicative that daclatasvir contributes to regimen efficacy also in the presence of polymorphisms at positions 30 and 93.

Genotypes 4, 5 and 6

Data from the AI444042 study, as well as a small sample from the AI444010 study, indicates that the efficacy of daclatasvir against genotype 4 is not lower than against genotype 1. In both these studies, daclatasvir was used in combination with pegIFN/RBV. There are no clinical data on the use of daclatasvir in genotypes 5 and 6, which are rare in Europe and the US. In vitro data are indicative that there will be relevant antiviral activity.

The efficacy of daclatasvir in cirrhotics

An important limitation in the available efficacy demonstration, is the absence of trial data for sofosbuvir+daclatasvir in patients with cirrhosis, with or without hepatic impairment. A small sample from the use of daclatasvir+asunaprevir in genotype 1b are indicative that daclatasvir, as a component of an interferon-free regimen, is capable of delivering high SVR rates in compensated cirrhotics. Furthermore, there are no safety issues or pharmacokinetic issues to preclude the use of daclatasvir in cirrhotics.

2.5.4. Conclusions on the clinical efficacy

Daclatasvir, when combined with sofosbuvir, is likely to provide a highly effective regimen in genotype 1 and by extrapolation also in genotype 4. Ribavirin is likely not needed for regimen optimisation in most patients with these genotypes. Data, however, are scarce for other genotypes than 1. Furthermore, the optimal duration of therapy is not well defined in many situations.

Data support the use of daclatasvir+sofosbuvir in genotype 1 and 4. Furthermore, daclatasvir has activity against genotypes 2 and 3 which is expected to be clinically relevant within appropriate regimens. These,

however, have not been defined. At present, the recommended use of daclatasvir in these genotypes is limited to patients with genotype 3 infection, cirrhosis and/or prior treatment experience, in whom available interferon free alternatives (sofosbuvir+ribavirin for 24 weeks) is anticipated to be associated with relapse in a significant proportion of patients. In this situation, the addition of daclatasvir to the regimen is considered appropriate.

2.6. Clinical safety

The safety database is primarily based on the assessment of 2 different DCV-combination regimens: DCV combined with the oral DAA sofosbuvir+/- ribavirin (RBV), and DCV combined with peginterferon alfa plus RBV (pegIFNα/RBV). Additional supportive safety information at the recommended dose of DCV is also presented for DCV in other combinations, including DCV combined with the BMS investigational NS3/4A protease inhibitor, asunaprevir (ASV).

Patient exposure

In support of the proposed indication, clinical safety data were provided from one pivotal study of DCV/SOF +/- RBV (AI444040; N = 211) and 6 randomized, double-blind, placebo controlled, supportive registrational Phase 2a/2b studies of DCV/pegIFNα/RBV (AI444010, AI444011, AI444014, AI444021, AI444022, and AI444031 N = 505). In addition, other supportive safety findings from 3 completed Phase 2/3 studies of DCV/ASV (AI447026, AI447017, and AI447011; N = 273) are also presented in the summary of clinical safety. Collectively, data across these 10 Phase 2/3 studies in 989 subjects exposed to DCV 60 mg QD support the application.

During the evaluation updated safety data was provided on subjects treated with DCV-combination regimens at recommended Dose (DCV 60 mg QD) in completed studies, for a total of 2,134 patients (see summary table).

The safety profile of daclatasvir is based on data from 798 patients with chronic HCV infection who received the daclatasvir 60 mg recommended daily dose either in combination with Sofosbuvir with or without ribavirin or in combination with peginterferon alfa and ribavirin (described in the SmPC section 4.8).

Summary of Subjects Treated with DCV-combination Regimens at Recommended Dose (DCV 60 mg QD) in Completed Studies

•	• Number of Subjects			
Study Number	**DCV/SOF ± RBV[a]**	**DCV/pegIFNα/RBV[a]**	**DCV/ASV ± *pegIFN/RBV*[a,b]**	**Total DCV**
Pivotal Study				
AI444040	211	---	---	**211**
Supportive / Registrational Studies				
AI444010	---	158	---	
AI444011	---	199 → 369	---	**369**
AI444014	---	12	---	
AI444031	---	100	---	**100**
AI444021	---	19	---	
AI444022	---	17 → 36	---	**36**
AI444042	---	***82***		***82***
Other Supportive Studies				
AI447028 [c]	---	---	***645***	***645***
AI447011 [c]	---	---	18 [c] ***20*** [d]	***38***
AI447017 [c]	---	---	33	
AI447026 [c]	---	---	222 → 255	**255**
AI447029 [d]	---	---	***398*** [d]	***398***
Total	**211**	***587***	***1,336***	***2,134***

Abbreviations: ASV - asunaprevir, DCV - daclatasvir, pegIFNα - pegylated interferon alfa, RBV - ribavirin, SOF - sofosbuvir

a Safety data from other DCV doses are not integrated in the overall by-regimen safety analyses.

b Safety data from DCV 60 mg QD in combination with a dose of ASV other than ASV 100 mg BID softgel capsule or ASV 200 mg BID (ASV at 600 mg BID and ASV at 200 mg QD) are not integrated in the overall by-regimen safety analyses.

c Subjects received DCV 60 mg QD in combination with ASV 100 mg BID softgel.

d Subjects received DCV 60 mg QD in combination with ASV/pegIFNα/RBV - DCV Quad.

The safety database for daclatasvir is considered sufficient for its evaluation within this MAA procedure. It is notable that daclatasvir has been studied in several different drug combinations, and has therefore been associated with adverse effects characteristic of several different co-treating agents.

Subjects with compensated cirrhosis at baseline were included in several studies evaluating DCV-containing regimens. In studies evaluating DCV 60 mg QD in combination with pegIFNα/RBV that enrolled cirrhotic subjects (AI444010, AI444011, and AI444031), 53 of 457 (11.6%) subjects had baseline cirrhosis. Of the 457 subjects enrolled, 400 were non-cirrhotic, 53 were cirrhotic, and 4 subjects were either missing or not reported at baseline (1 missing in AI444010, 3 not reported in AI444031). In

the study evaluating DCV 60 mg QD in combination with ASV (AI447026), 22 of 222 (10.0%) subjects had baseline cirrhosis. Notably, the safety database in cirrhotic patients is small.

All comparative safety data with daclatasvir were generated as an add-on to peginterferon and ribavirin, in comparison to pegIFN/RBV alone. The proportion of patients discontinuing due to adverse events was lower in those receiving triple therapy than those receiving only the bitherapy background. The proportion of patients discontinuing due to AEs in the pegIFN/RBV control arms was in the anticipated range, based on previous clinical trial experiences.

Adverse events

It is notable that daclatasvir was not associated with an increase in severe or serious AEs, or discontinuations due to AEs, compared to the background. Overall, these data are indicative of a drug that is well tolerated over the relevant treatment duration.

Overview of adverse events on treatment by DCV-combination regimen

	Number of Subjects (%)				
		DCV/pegIFNα/RBV[b]		Total[c]	
	DCV/SOF ± RBV[a] (N = 211)	DCV/pegIFNα/RBV (N = 505)	PBO/pegIFNα/RBV (N = 174)	DCV (N = 716)	DCV/ASV[d] (N = 273)
Adverse Events					
Deaths	0	0	0	0	0
Overall AEs	189 (89.6)	500 (99.0)	170 (97.7)	689 (96.2)	239 (87.5)
Treatment-related AEs	130 (61.6)	487 (96.4)	165 (94.8)	617 (86.2)	173 (63.4)
Grade 3/4 treatment-related AEs	0	80 (15.8)	44 (25.3)	80 (11.2)	32 (11.7)
Treatment-related SAEs	4 (1.9)	14 (2.8)	4 (2.3)	18 (2.5)	6 (2.2)
Treatment-related AEs leading to discontinuation	0	26 (5.1)	11 (6.3)	26 (3.6)	13 (4.8)

Abbreviations: AEs - adverse events; ASV - asunaprevir; DCV - daclatasvir; PBO - placebo; pegIFNα/RBV - peginterferon α plus ribavirin; RBV - ribavirin;
SAEs - searious adverse events; SOF - sofosbuvir
a DCV/SOF study: AI444040
b DCV/pegIFNα/RBV studies: AI444010, AI444011, AI444014, AI444021, AI444022, AI444031
c DCV/SOF and DCV/pegIFNα/RBV studies: AI444040, AI444010, AI444011, AI444014, AI444021, AI444022, AI444031
d DCV/ASV: AI447026, AI447017, and AI447011

Daclatasvir+sofosbuvir +/- ribavirin

The most common treatment emergent adverse events reported with sofosbuvir+daclatasvir are fatigue, headache and nausea. Anemia was exclusively reported when ribavirin was in the regimen. Further ribavirin-associated side effects include pruritus, cough, dyspnea and rash. All in all, no signature side effect profile of daclatasvir emerges in this study.

Summary Adverse Events On Treatment Prior to Addition of Rescue Therapy: Grouped by Treatment and Duration (AI444040) - Treated Subjects

	Number (%) of Subjects				
	DCV/SOF With RBV[a]		DCV/SOF Without RBV[b]		Total (N = 211)
	12 Weeks (N = 41)	24 Weeks (N = 49)	12 Weeks (N = 41)	24 Weeks (N = 80)	
Deaths	0	0	0	0	0
SAEs (Any Grade)	1 (2.4)	6 (12.2)	1 (2.4)	7 (8.8)	15 (7.1)
Treatment-related SAEs	1 (2.4)[c]	3 (6.1)[c]	0	0	4 (1.9)
AEs Leading to Discontinuation	0	1 (2.0)	0	1 (1.3)	2 (0.9)
Overall AEs (Any Grade)	38 (92.7)	46 (93.9)	38 (92.7)	67 (83.8)	189 (89.6)
Grade 3/4 AEs	1 (2.4)	3 (6.1)	1 (2.4)	2 (2.5)	7 (3.3)
Treatment-related AEs (Any Grade)	26 (63.4)	40 (81.6)	22 (53.7)	42 (52.5)	130 (61.6)
Grade 3/4 treatment-related AEs	0	0	0	0	0
Treatment-related AEs (Any Grade ≥ 5% total)					
Fatigue	13 (31.7)	15 (30.6)	13 (31.7)	20 (25.0)	61 (28.9)
Headache	6 (14.6)	13 (26.5)	6 (14.6)	15 (18.8)	40 (19.0)
Nausea	5 (12.2)	10 (20.4)	7 (17.1)	8 (10.0)	30 (14.2)
Diarrhea	2 (4.9)	5 (10.2)	1 (2.4)	6 (7.5)	14 (6.6)
Anemia	7 (17.1)	6 (12.2)	0	0	13 (6.2)
Insomnia	1 (2.4)	6 (12.2)	2 (4.9)	3 (3.8)	12 (5.7)
Pruritus	5 (12.2)	4 (8.2)	0	3 (3.8)	12 (5.7)
AEs Commonly Associated with RBV					
Anemia	7 (17.1)	6 (12.2)	0	0	13 (6.2)
Cough	6 (14.6)	10 (20.4)	2 (4.9%)	1 (1.3)	19 (9.0)
Insomnia	4 (9.8)	7 (14.3)	4 (9.8)	4 (5.0)	19 (9.0)
Anxiety	2 (4.9)	5 (10.2)	0	4 (5.0)	11 (5.2)
Dyspnea	6 (14.6)	3 (6.1)	0	2 (2.5)	11 (5.2)
Rash (composite)	4 (9.8)	8 (16.3)	2 (4.1)	5 (6.3)	19 (9.0)

Abbreviations: AEs - adverse events, DCV daclatasvir, SAEs - serious adverse events, RBV - ribavirin, SOF - sofosbuvir
a Group H: 12 weeks; Groups E, F and J: 24 weeks
b Group G: 12 weeks; Groups A, B, C, D, and I: 24 weeks
c These events of overdose were generally inadvertent single extra doses of study medications reported as SAEs per protocol and did not result in clinical symptoms or require intervention or treatment.

Daclatasvir+peg/IFN/RBV

The side effect profile of peginterferon+ribavirin is well described and included haematological side effects, neuropsychiatric effects, influenza-like illness, thyroid disorders and the possibility of precipitating autoimmune disease. Furthermore, interferons are ill tolerated in patients with advanced liver disease, where it may precipitate serious bacterial infections and probably also hepatic decompensation.

Summary Adverse Events On Treatment DCV/pegIFNα/RBV (Recommended Dose) in AI444010, AI444011, AI444014, AI444031, AI444021, and AI444022 and AI444042 – Treated Subjects

	Number (%) of Subjects	
	DCV/ pegIFNα/RBV[a] (N= 587)	Placebo/pegIFNα/RBV[b] (N= 216)
Deaths	0	0
SAEs (any grade)	33 (5.6)	14 (6.5)
Treatment related SAEs (any grade)	16 (2.7)	5 (2.3)
AEs leading to discontunuation	36 (6.1)	18 (8.3)
Overall AEs (any grade)	580 (98.8)	210 (97.2)
Treatment related AEs (any grade)	107 (18.2)	58 (26.9)
Grade 3&4 Treatment related AEs	566 (96.4)	205 (94.9)
Treatment related AEs (any grade ≥ 5% total)	94 (16.0)	52 (24.1)
Fatigue	225 (38.3)	95 (44.0)
Headache	191 (32.5)	71 (32.9)
Pruritus	184 (31.3)	60 (27.8)
Insomnia	149 (25.4)	63 (29.2)
Influenza like illness	149 (25.4)	40 (18.5)
Dryn slin	128 (21.8)	31 (14.4)
Alopecia	117 (19.9)	32 (14.8)
Nausea	126 (21.5)	41 (19.0)
Decreased appetite	124 (21.1)	43 (19.9)
Rash	113 (19.3)	51 (23.6)
Asthenia	112 (19.1)	38 (17.6)
Irritability	106 (18.1)	42 (19.4)
Myalgia	103 (17.5)	53 (24.5
Anemia	95 (16.2)	47 (21.8)
Pyrexia	86 (14.7)	39 (18.1)
Cough	83 (14.1)	36 (16.7)
Dyspnea	82 (14.0)	30 (13.9)
Neutropenia	76 (12.9)	43 (19.9)
Diarrhea	71 (12.1)	23 (10.6)
Arthralgia	71 (12.1)	37 (17.1)
Depression	56 (9.5)	26 (12.0)
Chills	51 (8.7)	27 (12.5)
Injection site erythema	39 (6.6)	9 (4.2)
Dizziness	38 (6.5)	17 (7.9)
Dyspnea exertionl	38 (6.5)	(4.2)
Dysgeusia	35 (6.0)	9 (4.2)
Anxiery	33 (5.6)	16 (7.4)
Back Pain	33 (5.6)	14 (6.5)
Vomiting	33 (5.6)	15 (6.9)
Sleep disorder	31 (5.3)	5 (2.3)
Injection site reaction	24 (4.1)	11 (5.1)
Disturnace in attention	23 (3.9)	11 (5.1)
Abdomianl pain	20 (3.4)	11 (5.1)
Dyspepsia	17 (2.9)	12 (5.6)
Weight increased	(2.9)	18 (8.3)
Thrombocypenl	16 (2.7)	11 (5.1)

Abbreviations: AEs - adverse events; DCV - daclatasvir; PBO - placebo; pegIFNα/RBV - peginterferon α plus ribavirin; RBV - ribavirin, SAEs - serious adverse events
a Includes subjects treated with DCV 60 mg QD/pegIFNα/RBV in AI444010, AI444011, AI444014, AI444031, AI444021, AI444022
b Includes subjects treated with placebo/pegIFNα/RBV in AI444010, AI444011, AI444014, AI444031, AI444021, AI444022 and AI444042

In summary, these data are not indicative that daclatasvir increases the frequency or severity of any particular side effect, or is associated with a general deterioration of the regimen side effect profile, when added to peginterferon and ribavirin bitherapy.

Serious adverse event/deaths/other significant events

Daclatasvir+sofosbuvir +/- ribavirin

There were no deaths reported in this study.

One subject experienced a SAE of cerebrovascular accident (CVA) and 1 subject experienced an AE of fibromyalgia. No subjects with a METAVIR score of F2 or greater required discontinuation of study therapy due to an AE. The 2 subjects who discontinued study therapy due to an AE had a calculated METAVIR score of F0 - F1.

One case of death due to cardiac failure in the context of septicaemia and hepatic decompensation was reported in the French compassionate use program. This prompted a thorough review of the cardiovascular safety of daclatasvir. No indication of cardiovascular toxicity was identified.

Daclatasvir in combination with pegIFN/RBV:

There were no deaths reported on treatment in subjects treated with DCV 60 mg QD/pegIFNα/RBV.

In AI444010, AI444011, and AI444014, 4 subjects treated with DCV 20 mg/pegIFNα/RBV died, either on study or during follow-up (unknown causes, hepatocellular carcinoma, intravenricular haemorrhage, cardiopulmonary failure/asthma. No clear pattern occurs in these cases. Furthermore, two of the four cases died during follow up rather than while exposed to daclatasvir.

Overall, the frequency of reported SAEs, regardless of relationship to study therapy, was similar among DCV/pegIFNα/RBV-treated subjects (29/505 [5.7%]) and placebo/pegIFNα/RBV-treated subjects (12/174 [6.9%]).

Laboratory findings

The impact of ribavirin on the haematological safety profile is apparent. In the absence of ribavirin there were no grade 3-4 haematological laboratory abnormalities.

Results from the AI444042 study are congruent with the above findings (no additive haematological toxicity), with the exception of an increase in grade 3-4 decreases in leukocytes/lymphocytes. This finding was not apparent on a full pooling of data from the placebo controlled studies where daclatasvir was given in combination with pegIFN/RBV.

The decrease in haemoglobin of around 2.5 g/dL is characteristic of ribavirin. The on-treatment decrease of haemoglobin in the daclatasvir+sofosbuvir arms without ribavirin is noted. The finding may be seen as somewhat surprising, as sofosbuvir has not been associated with haematological side effects and, as seen below, daclatasvir does not seem to aggravate pegIFN/RBV associated anemia. Furthermore, it is noted that this effect was also seen during the first week when sofosbuvir was given as monotherapy in the lead in-phase. The applicant has proposed that the intensive blood sampling protocol at the initiation of the study may be responsible for this decline. The magnitude of the effect is not considered clinically relevant.

Worst Grade of On Treatment Hematologic Laboratory Abnormalities in AI444040 - Treated Subjects

	DCV/SOF ± RBV[c] (N=211) Number (%) of Subjects		
Parameter	DCV/SOF With RBV N = 90	DCV/SOF Without RBV N = 121	Total N = 211
Hemoglobin			
Grade 0	62 (68.9)	117 (96.7)	179 (84.8)
Grade 1-2	27 (30.0)	4 (3.3)	31 (14.7)
Grade 3-4	1 (1.1)	0	1 (0.5)
Platelet count			
Grade 0	88 (97.8)	113 (93.4)	201 (95.3)
Grade 1-2	2 (2.2)	8 (6.6)	10 (4.7)
Grade 3-4	0	0	0
Neutrophils			
Grade 0	88 (97.8)	118 (97.5)	206 (97.6)
Grade 1-2	2 (2.2)	3 (2.5)	5 (2.4)
Grade 3-4	0	0	0

c Data are presented prior to the addition of rescue therapy

Worst Grade of On Treatment Liver Function Laboratory Abnormalities in AI444040 - Treated Subjects

	DCV/SOF[c] (N=211) Number (%) of Subjects		
Parameter	DCV/SOF with RBV N = 90	DCV/SOF without RBV N = 121	Total N = 211
ALT			
Grade 0	82 (91.1)	109 (90.0)	191 (90.5)
Grade 1-2	8 (8.9)	12 (9.9)	20 (9.5)
Grade 3-4	0	0	0
AST			
Grade 0	83 (92.2)	106 (87.6)	189 (89.6)
Grade 1-2	7 (7.7)	15 (12.4)	22 (10.4)
Grade 3-4	0	0	0
Total Bilirubin			
Grade 0	73 (81.1)	115 (95.0)	188 (89.1)
Grade 1-2	17 (18.9)	6 (5.0)	23 (10.9)
Grade 3-4	0	0	0

c Data are presented prior to the addition of rescue therapy.

There is no signal of potential hepatotoxicity with concomitant use of sofosbuvir+daclatasvir.

Safety in special populations

Subjects were required to be non-cirrhotic at baseline per the study protocol in study AI444040.

Subjects with baseline compensated cirrhosis were included in several studies evaluating DCV-containing regimens. In studies evaluating DCV 60 mg QD in combination with pegIFNα/RBV that enrolled cirrhotic subjects (AI444010, AI444011, and AI444031), of the 457 subjects enrolled, 400 were non-cirrhotic, 53 were cirrhotic, and 4 subjects were either missing or not reported at baseline (1 missing in AI444010, 3 not reported in AI444031).

Treatment-related Adverse Events Reported in at Least 20% of DCV/pegIFNα/RBV-treated Subjects by Cirrhosis Status (Cirrhosis or No Cirrhosis)

	Number of Subjects (%)[a]			
	Cirrhosis		No Cirrhosis	
Preferred Term	DCV/ pegIFNα/RBV N = 53	PBO /pegIFNα/RBV N = 19	DCV /pegIFNα/RBV N = [illegible]	PBO /pegIFNα/RBV N = 125
Total subject with an event	50 (94.3)	18 (94.7)	[illegible]	117 (93.6)
Pruritus	22 (41.5)	4 (21.1)	[illegible]	36 (28.8)
Fatigue	16 (30.2)	7 (36.8)	189 (47.3)	65 (52.0)
Influenza-like illness	16 (30.2)	8 (42.1)	105 (26.3)	14 (11.2)
Insomnia	9 (17.0)	6 [illegible]	108 (27.0)	42 (33.6)
Asthenia	14 (26.4)	[illegible]	45 (11.3)	10 (8.0)
Dry skin	12 (22.6)	[illegible] (10.5)	101 (25.3)	20 (16.0)
Headache	12 (22.6)	5 (26.3)	137 (34.3)	43 (34.4)
Irritability	11 (20.8)	7 (36.8)	79 (19.8)	24 (19.2)
Nausea	11 [illegible]	6 (31.6)	96 (24.0)	20 (16.0)
Dyspnea	11 [illegible].8)	2 (10.5)	62 (15.5)	17 (13.6)
Myalgia	11 (20.8)	4 (21.1)	73 (18.3)	32 (25.6)
Alopecia	9 (17.0)	2 (10.5)	80 (20.0)	16 (12.8)

Abbreviations: DCV - daclatasvir, PBO - placebo, pegIFNα - pegylated interferon alfa, RBV - ribavirin
a Does not include AEs that may have occurred during rescue therapy.

Treatment-related Adverse Events Reported in at Least 20% of DCV/pegIFNα/RBV-treated Subjects by Cirrhosis Status (Cirrhosis or No Cirrhosis)

Preferred Term	Number of Subjects (%)[a]			
	Cirrhosis		No Cirrhosis	
	DCV/ pegIFNα/RBV N = 53	PBO /pegIFNα/RBV N = 19	DCV /pegIFNα/RBV N = 400	PBO /pegIFNα/RBV N = 125
Total subject with an event	50 (94.3)	18 (94.7)	385 (96.3)	117 (93.6)
Pruritus	22 (41.5)	4 (21.1)	125 (31.3)	36 (28.8)
Fatigue	16 (30.2)	7 (36.8)	189 (47.3)	65 (52.0)
Influenza-like illness	16 (30.2)	8 (42.1)	105 (26.3)	14 (11.2)
Insomnia	9 (17.0)	6 (31.6)	108 (27.0)	42 (33.6)
Asthenia	14 (26.4)	1 (5.3)	45 (11.3)	10 (8.0)
Dry skin	12 (22.6)	2 (10.5)	101 (25.3)	20 (16.0)
Headache	12 (22.6)	5 (26.3)	137 (34.3)	43 (34.4)
Irritability	11 (20.8)	7 (36.8)	79 (19.8)	24 (19.2)
Nausea	11 (20.8)	6 (31.6)	96 (24.0)	20 (16.0)
Dyspnea	11 (20.8)	2 (10.5)	62 (15.5)	17 (13.6)
Myalgia	11 (20.8)	4 (21.1)	73 (18.3)	32 (25.6)
Alopecia	9 (17.0)	2 (10.5)	80 (20.0)	16 (12.8)

Abbreviations: DCV - daclatasvir, PBO - placebo, pegIFNα - pegylated interferon alfa, RBV - ribavirin
a Does not include AEs that may have occurred during rescue therapy.

Liver Function Test Laboratory Abnormalities Reported in DCV/pegIFNα/RBV-treated Subjects by Cirrhosis Status (Cirrhosis or No Cirrhosis)

	Number of Subjects (%)[a,b]			
	Cirrhosis		No Cirrhosis	
Preferred Term	DCV/ pegIFNα/RBV N = 53	PBO /pegIFNα/RBV N = 19	DCV /pegIFNα/RBV N = 400	PBO /pegIFNα/RBV N = 125
ALT	N = 53	N = 18	N = 399	N = 123
Grade 0	20 (37.7)	5 (27.8)	258 (64.7)	59 (48.0)
Grade 1-4	33 (62.3)	13 (72.2)	141 (35.3)	64 (52.0)
Grade 3-4	1 (1.9)	1 (5.6)	9 (2.3)	[illegible]
AST	N = 53	N = 18	N = 399	N = 123
Grade 0	12 (22.6)	3 (16.7)	242 (60.7)	[illegible] (46.3)
Grade 1-4	41 (77.4)	15 (83.3)	157 (39.3)	66 (53.7)
Grade 3-4	3 (5.7)	2 (11.1)	9 (2.3)	1 (0.8)
Total Bilirubin	N = 53	N = 18	N = 399	N = 123
Grade 0	34 (64.2)	16 (88.9)	[illegible] (81.7)	94 (76.4)
Grade 1-4	19 (35.8)	2 (11.1)	[illegible] (18.3)	29 (23.6)
Grade 3-4	2 (3.8)	1 (5.6)	2 (0.5)	2 (1.6)

Abbreviations: DCV - daclatasvir, PBO - placebo, pegIFNα - pegylated interferon alfa, RBV - ribavirin
a Does not include assessments that may have occurred during rescue therapy.
b Percentage relative to the number of subjects with laboratory test results.

Rates of drug-related AEs were similar in subjects treated with DCV/ASV with and without baseline cirrhosis in study AI447026 (59.1% [13/22] vs 57.5% [115/200], respectively).

Treatment-related Adverse Events Reported in at Least 5% DCV/ASV-treated Subjects in AI447026 by Cirrhosis Status (Cirrhosis or No Cirrhosis)

	Number of Subjects (%)[a,b]	
	Cirrhosis	No Cirrhosis
Preferred Term	N = 22	N = 200
Total subject with an event	13 (59.1)	115 (57.5)
ALT increased	2 (9.1)	33 (16.5)
Headache	1 (4.5)	21 (10.5)
AST increased	1 (4.5)	27 (13.5)
Pyrexia	3 (13.6)	21 (10.5)
Diarrhea	1 (4.5)	13 (6.5)
Eosinophilia	0	11 (5.5)
Malaise	2 (9.1)	5 (2.5)
Bronchitis	2 (9.1)	8 (4.0)

Abbreviations: ALT - alanine aminotransferase, AST - aspartate aminotransferase
a Does not include AEs that may have occurred during rescue therapy.
b Subjects were not pooled across ineligible-naive/intolerant and prior non-responder populations.

The safety database in cirrhotic patients is small. However, available data are not indicative of any deterioration of the safety profile of daclatasvir when cirrhosis is present. There is no increase in exposure in advanced liver disease, and no side effects have been identified in the general population that would be anticipated to be more severe in patients with advanced liver disease.

2.6.1. Discussion on clinical safety

The primary safety database submitted for this application contains 989 patients treated with daclatasvir+sofosbuvir+/-ribavirin (n=211, no definite cirrhotics), daclatasvir+PegIFN/RBV or daclatasvir+asunaprevir and 75 of these had cirrhosis. During the evaluation updated safety data was provided on subjects treated with DCV-combination regimens at recommended Dose (DCV 60 mg QD) in completed studies. The safety profile of daclatasvir is based on data from 798 patients with chronic HCV infection who received the daclatasvir 60 mg recommended daily dose either in combination with sofosbuvir with or without ribavirin or in combination with peginterferon alfa and ribavirin. The emerging side effects profile does not clearly differ from placebo. In comparative studies as an add-on versus placebo to pegIFN+ribavirin, there is no increase in side effects. In the absence of ribavirin, there appears to be no reasonable evidence that any particular side effect is causally related to daclatasvir.

The relatively small database on safety in cirrhotics is recognised, as is the near-absence of data in patients with decompensated liver disease/hepatic impairment. However it is notable that no side effects that would be anticipated to be more severe in cirrhotics have been identified in the general population. Furthermore, exposure to the active moiety of daclatasvir was not impacted by Child-Pugh stage in a hepatic impairment study. Therefore, there are no specific safety concerns relevant to the use of daclatasvir in patients with advanced liver disease.

All the adverse reactions reported in clinical trials have been included in the Summary of Product Characteristics.

2.6.2. Conclusions on the clinical safety

While the safety database in cirrhotic patients is limited, and there is little systematic experience in patients with hepatic impairment, the general safety profile of daclatasvir does not clearly differ from placebo. Furthermore, exposure to unbound daclatasvir is not altered in advanced liver disease. There are no specific safety concerns to preclude exposure to daclatasvir in patients in need of antiviral therapy to achieve HCV clearance.

2.7. Pharmacovigilance

Detailed description of the pharmacovigilance system

The CHMP considered that the Pharmacovigilance system as described by the applicant fulfils the legislative requirements.

2.8. Risk Management Plan

The CHMP received the following PRAC Advice on the submitted Risk Management Plan:

PRAC Advice

Based on the PRAC review of the RMP version 1.2, the PRAC considers by consensus that the risk management system for daclatasvir (Daklinza), in combination with other agents, in the treatment of chronic hepatitis C (CHC) in adults is acceptable.

This advice is based on the following content of the Risk Management Plan:

- **Safety concerns**

Summary of safety concerns	
Important identified risks	*None*
Important potential risks	- CYP3A inhibitors and inducers; P-gp i-inhibitors, inducers, and substrates; OATP1B1, OATP1B3, and BCRP substrates. - Hepatic Toxicity; - Hematologic Toxicity; - Development of Drug Resistance; - Embryo-fetal Development Toxicity.
Missing information	- Pregnancy and Lactation; - Children and Adolescents (<18 years of age); - HIV/HCV; - HBV/HCV; - Hepatic Impairment and Decompensated Liver Disease; - Liver Transplant; - Subjects aged > 65 years; - Subjects of African origin; - Subjects co-medicated with interacting agents dosed at either 30 mg/day or 90 mg/day.

- **Pharmacovigilance plans**

Ongoing and Planned Additional Pharmacovigilance Studies in the Pharmacovigilance Plan

Study (type and study number)	**Safety concern addressed**	**Planned date for submission of interim or final reports**
Ongoing studies		
AI444038: Phase 3 nonrandomized, open-label, study of DCV/pegIFNα/RBV in GT-1 treatment-naïve African American, Hispanic/Latino, and Caucasian subjects	Race/Ethnicity assessment. (~230 subjects)	Final CSR submission April 2015
AI444043: Phase 3 nonrandomized, open-label study of DCV/pegIFNα/RBV in GT-1 treatment-naïve subjects co-infected with HIV	The use of DCV in HIV/HCV co-infected individual has not been established. (~300 subjects)	Final CSR submission April 2015
AI444046: Phase 3 nonrandomized, open-label, long-term follow-up and observational study of durability of efficacy, resistance, and characterization of progression of liver disease in subjects with CHC previously treated with DCV and/or ASV	Durability of DCV clinical benefit in large, observational study of subjects previously treated with DCVcontaining regimen. (~1000 subjects)	Final CSR submission 4Q/2019
AI444215: Phase 3 study of DCV/sofosbuvir in subjects with cirrhosis who may require future liver transplant and subjects post-liver transplant (both cases for GT 1-6)	The use of DCV in patients with liver transplant has not been established.	Final CSR submission July 2015
AI444217: Phase 3 study of DCV/sofosbuvir in subjects coinfected with HIV and previously untreated (GT 1-6)	The use of DCV in HIV/HCV co-infected individual has not been established.	Final CSR submission July 2015
AI444273: A Phase 1, open-label, crossover study to evaluate the drug interaction between dolutegravir and DCV in healthy adult subjects	The impact of co-administration of these agents on PK has not been established.	Final CSR submission April 2015
Planned studies		
AI444093: A Phase 1 Clinical Study to Assess the Effect of Darunavir/Ritonavir or Lopinavir/Ritonavir on the Pharmacokinetics of Daclatasvir in Healthy Subjects	The impact of DCV dose adjustment due to drug interactions has not been establihed	Final CSR submission April 2015
Paediatric Studies: A paediatric investigational plan (PIP number	The use of DCV in paediatric patients has not been	To Be Determined (all clinical studies are deferred)

Study (type and study number)	Safety concern addressed	Planned date for submission of interim or final reports
EMEA-001191-PIP01- 11) has been proposed and agreed by the EMA in 2012 (Decision number P/0166/2012)	established.	
In vitro study with DCV using a human hepatocyte model, and possibly cells expressing individual uptake transporter, to evaluate the involvement of transporters, including OCT1, in the hepato-bilary excretion of DCV	Active transport may contribute to the hepato-bilary excretion of DCV and be a source of PK variability	1Q2015

DCV = daclatasvir, ASV = Asunaprevir

- **Risk minimisation measures**

Summary of Risk Minimization Measures		
Safety Concern	**Risk Minimization Measures**	
	Routine	**Additional**
Drug-drug Interaction	The following guidance is provided in the SmPC: • SmPC section 4.2 Posology:Dose recommendation for concomitant medicines • SmPC section 4.4 Warnings: Interactions with medicinal products. • DCV is contraindicated when combined with medicinal products that strongly induce CYP3A4 and P-gp (SmPC section 4.3). • SmPC section 4.5 provides for established and other potentially significant drug-drug interactions. • Use caution: Digoxin, Rosuvastatin and other substrates of OATP1B1 and BCRP • Dose adjustment guidance: Strong inhibitors of CYP3A4: the dose of DCV should be reduced to 30 mg QD. Moderate inducers of CYP3A4: the dose of DCV should be increased to 90 mg once daily.	None
Hepatic Toxicity	SmPC includes the warning/precaution that the safety and efficacy of DCV has not been established in patients with decompensated liver disease.	None
Hematologic Toxicity	Routine PhV	None
Development of Drug Resistance	SmPC includes the warning/precaution that DCV must not be administered as monotherapy. Also, in the posology section monitoring of HCV RNA levels during treatment is recommended in the SmPC, with discontinuation of therapy recommended for patients treated with DCV and pegIFNα/RBV experiencing confirmed virologic breakthrough (treatment stopping rules provided for weeks 4, 12 and 24).	None
Embryo-fetal Development Toxicity	SmPC section 4.6 (Pregnancy and lactation) states that DCV should not be used during pregnancy or in women of childbearing potential not using contraception. Use of highly effective contraception should be continued for 5 weeks after completion of DCV therapy (SmPC section 4.6). Since DCV in combination with pegIFNα/RBV is one of the recommended regimens in the SmPC, section 4.4 (Special warnings and precautions for use) states: When DCV is used in combination with ribavirin, the contraindications and warnings applicable to that medicinal product are applicable. Significant teratogenic and/or embryocidal effects have been demonstrated in all animal species exposed to ribavirin; therefore, extreme care must be taken to avoid pregnancy in female patients and in female partners of male patients (see the Summary of Product	None

Summary of Risk Minimization Measures		
	Characteristics for ribavirin).	
Pregnancy and Lactation	SmPC section 4.6 (Pregnancy and lactation) states that DCV should not be used during pregnancy or in women of childbearing potential not using contraception. Use of highly effective contraception should be continued for 5 weeks after completion of DCV therapy (SmPC section 4.6). Since DCV in combination with pegIFNα/RBV is one of the recommended regimens in the SmPC, section 4.4 (Special warnings and precautions for use) states: When DCV is used in combination with ribavirin, the contraindications and warnings applicable to that medicinal product are applicable. Significant teratogenic and/or embryocidal effects have been demonstrated in all animal species exposed to ribavirin; therefore, extreme care must be taken to avoid pregnancy in female patients and in female partners of male patients (see the Summary of Product Characteristics for ribavirin).	None
Children and Adolescents (18 years of age)	SmPC 4.4 under Special warnings and precautions for use: The safety and efficacy of DCV in the treatment of HCV infection in children and adolescents aged below 18 years have not been established.	None
HIV/HCV Co-infection	SmPC 4.4 under Special warnings and precautions for use: The safety and efficacy of DCV in the treatment of HCV infection in patients who are co-infected with HIV have not been established.	None
HBV/HCV Co-infection	SmPC 4.4 under Special warnings and precautions for use: The safety and efficacy of DCV in the treatment of HCV infection in patients who are co-infected with HBV have not been investigated.	None
Hepatic Impairment and Decompensated Liver Disease	SmPC 4.4 under Special warnings and precautions for use: The safety and efficacy of DCV in the treatment of HCV infection in patients with decompensated liver disease have not been established.	None
Liver transplant	SmPC 4.4 under Special warnings and precautions for use: The safety and efficacy of DCV in the treatment of HCV infection in patients who are pre-, peri-, or post-liver transplant or other organ transplant patients have not been established.	None
African Origin	Routine PhV. As per clinical guidance, HCV RNA levels should be monitored during treatment for patients receiving DCV with pegIFNα/RBV. Study AI444038 ongoing.	None
Age > 65 Years	Routine PhV, SmPC 4.4. Clinical data in patients aged 65 years and older are limited.	None
Subjects in whom drugs with potential for clinically significant DDI may be expected to decrease systemic exposure to DCV	SmPC section 4.4 Warnings: Interactions with medicinal products. SmPC section 4.5 (Interaction with other medicinal products and other forms of interaction). Studies AI444043 and AI444216 are ongoing. Routine PhV.	None

DCV = daclatasvir

The CHMP endorsed this advice without changes.

2.9. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the applicant show that the package leaflet meets the criteria for readability as set out in the *Guideline on the readability of the label and package leaflet of medicinal products for human use.*

The applicant has submitted an acceptable bridging statement regarding the lower strength not subject to user consultation.

3. Benefit-Risk Balance

Benefits

Beneficial effects

In the single pivotal trial part of this application, daclatasvir was used in combination with sofosbuvir, with or without ribavirin, for 12 or 24 weeks, in non-cirrhotic treatment naïve patients with genotype 1, 2 or 3 HCV infection, and in non-cirrhotic patients with genotype 1 HCV infection that have previously experienced virological failure when treated with telaprevir or boceprevir in combination with pegIFN/RBV. Among 126 treatment naïve patients with genotype 1 treated for 12 or 24 weeks, 124 achieved SVR12 (98.4%). There was no incremental effect of adding ribavirin to daclatasvir+sofosbuvir and there was no incremental effect of 12 more weeks of therapy after the first three months.

In 41 patients with genotype 1 infection that had prior virological failure on an NS3/4A inhibitor in combination with pegIFN/RBV and were treated with daclatasvir+sofosbuvir for 24 weeks with or without ribavirin, 40 achieved a documented SVR12 (97.6%). There was no apparent benefit of adding ribavirin.

Among 44 patients with genotypes 2 or 3 HCV infection (26 with genotype 2, 18 with genotype 3), 40 patients reached documented SVR12 (90.9%). One patient with genotype 3 HCV infection was termed a virological failure due to virological breakthrough and received rescue medication. However, this patient would not have been considered a virological failure, and rescue medication would not have been mandated according to current criteria. One patient with genotype 3 infection and with a baseline viral polymorphism reducing susceptibility to daclatasvir experienced relapse.

In a comparative study of daclatasvir + pegIFN/RBV for 24-48 weeks versus placebo + pegIFN/RBV for 48 weeks in treatment-naïve patients with genotype 4 infection, 125 patients were randomised 2:1 to either arm. SVR rates were 81.7% in the daclatasvir arm compared to 42.9% in the placebo arm. The difference was 38.8% ($p < 0.00001$).

Uncertainty in the knowledge about the beneficial effects

While there are clear indications from the study program where daclatasvir was used in combination with pegIFN/RBV, that there is a correlation between in vitro EC50 values and the clinical efficacy of daclatasvir, this was not seen when daclatasvir is used in combination with sofosbuvir. While there is evidence of the contribution of daclatasvir to regimen efficacy also in situations where the EC50 is a 1000-fold higher than that seen in genotype 1, it is unclear at what in vitro susceptibility no clinically relevant effect of daclatasvir would be expected in different treatment situations relevant to the use of daclatasvir+sofosbuvir.

It is unclear to what extent daclatasvir would contribute to the activity of a retreatment regimen after non-curative exposure to an NS5A inhibitor.

There are no data on the efficacy of sofosbuvir+daclatasvir in patients with cirrhosis. The optimal treatment duration is unknown in patients with genotype 1 infection and advanced liver disease. Furthermore, it remains unknown whether adding ribavirin is beneficial in such patients.

It is notable that the viral susceptibility to daclatasvir is lower in genotypes 2 and 3, compared to -1 and -4. The database for the use of sofosbuvir+daclatasvir in genotypes 2 and 3 is too small for a precise efficacy measure; furthermore, as opposed to the case with genotype 1, no patients known to be “difficult to treat” have been included in the available studies. While there is antiviral activity which is likely to be clinically relevant, the appropriate treatment duration with sofosbuvir+daclatasvir has not been determined, nor can the contribution of ribavirin be precisely evaluated.

Risks

Unfavourable effects

Daclatasvir has been studied extensively in combination with a number of different drugs. The primary safety database for this application contains 989 patients treated with daclatasvir+sofosbuvir+/-ribavirin, daclatasvir+PegIFN/RBV or daclatasvir+asunaprevir and 75 of these had cirrhosis. The emerging side effects profile does not clearly differ from placebo. Updated safety data was provided on subjects treated with DCV-combination regimens at recommended Dose (DCV 60 mg QD) in completed studies. The updated safety database contains 2134 patients exposed to daclatasvir, of which 798 patients received the daclatasvir 60 mg recommended daily dose either in combination with sofosbuvir with or without ribavirin or in combination with peginterferon alfa and ribavirin. The most frequently adverse reactions observed with daclatasvir in combination were headache, nausea and fatigue.

In comparative studies as an add-on versus placebo to pegIFN+ribavirin, there is no increase in side effects. There appears to be no reasonable evidence that any particular side effect is causally related to daclatasvir.

Uncertainty in the knowledge about the unfavourable effects

The safety database in patients with cirrhosis is relatively small. There is limited safety data in patients with hepatic impairment/decompensated liver disease.

Benefit-risk balance

Effects Table for daclatasvir

	Effect	Short Description	Unit	Daclatasvir+ sofosofosbuvir +/- ribavirin	Daclatasvir + pegIFN+ribaviri n	Uncertainties/ Strength of evidence	References
Favourable	SVR	Plasma HCV-RNA <LLOQ 12 weeks post planned end of therapy	%	9[illegible] genotypes 1,2 and 3	Genotype 4: 82 with daclatasvir +pegIFN/RBV compared to 43 with placebo + pegIFN/RBV. Difference +39 (p <0.00001)	Efficacy in genotype 1 very high also in difficult to treat patients. Strong evidence of high efficacy in genotype 1. Small sample in genotypes 2 and 3 with anticipated high background regimen efficacy; likely contribution to regimen efficacy but low evidence of precise effect. Data from genotype 4 are indicative of similar efficacy of daclatasvir in genotype 1 and -4	See discussion on clinical efficacy.
Unfavourable	Teratogenic potential Frequently observed adverse reactions with daclatasvir in combination were headache, nausea and fatigue.					Study did not include patients with hepatic impairment	See discussion on non-clinical and clinical safety.

Abbreviations: LLOQ=lower limit of quantification

Benefit-risk balance

Discussion on the benefit-risk balance

Daclatasvir was studied in combination with sofosbuvir, with or without ribavirin, in the AI444040 trial, nominally a phase IIb study. This pivotal study of the present application was a cross-company collaboration where the combination of two agents developed by different sponsors. Notwithstanding the outcomes of this trial, the development of sofosbuvir+daclatasvir was not continued into a phase III program, for industrial reasons.

The pivotal trial included non-cirrhotic patients infected with genotype 1, 2 or 3 virus. Apart from treatment naïve patients, the study included 41 patients with prior virological failure on an NS3/4A inhibitors+pegIFN/RBV. Results were outstanding in all treatment categories, with two nominal virological failures among a total of 211 patients. The criteria for an approval on the basis of one pivotal trial are considered to be met.

The group of patients with prior failure on NS3/4A inhibitor based therapy represent an important unmet medical need, as the efficacy of presently approved regimens is questionable in such patients. They also constitute a group of demonstrably “difficult to treat” patients demonstrating that the impressive virological efficacy seen in genotype 1 was not due to the selection of easy-to-treat patients. This is of importance, as the field of HCV drug development has seen cases where SVR rates in phase III were considerably lower than anticipated based on phase II results, presumably due to the selection of patients.

The safety profile of daclatasvir is not clearly distinct from placebo, and PK data in hepatic impairment are not indicative of increased exposure or the need for dose adjustment. Furthermore, available data on the use of daclatasvir in patients with cirrhosis, though limited, are promising.

Rationale for regimen recommendations

As available data on the efficacy of the daclatasvir+sofosbuvir combination is limited to relatively small study with different viral genotypes, the optimal treatment duration and the potential benefit of adding ribavirin to the regimen is not well-characterised. The reported clinical trial experience of daclatasvir use in cirrhotic patients pertains to other drug combinations, such as daclatasvir+peginterferon+ribavirin and daclatasvir+asunaprevir. While these are indicative that daclatasvir is effective in cirrhotic patients, they do not give any clue as to the optimal treatment duration with sofosbuvir+daclatasvir in cirrhotics, or to the possible value of adding ribavirin to such a regimen.

The study of treatment experienced patients is limited to patients with genotype 1 infection previously failing on a combination of a NS3/4A inhibitor + peginterferon/ribavirin. It is recognised that as no potential cross-resistance between previously used and presently planned drugs has been selected, a treatment experienced population is functionally to be considered a select subset of the more difficult to treat proportion of a treatment naïve population, e.g., having a higher mean age, less likely to have low baseline HCV-RNA, more likely to have more advanced fibrosis, and considerably more likely to have IL28B non C/C genotype, which negatively impacts the interferon response of the host.

It is notable, however, that due to the very high response rates seen in the studied population in AI444040, it is not possible to estimate to what extent such previously characterised negative prognostic factors impact the required treatment duration and the need for ribavirin, in order to maximise the probability of SVR with sofosbuvir+daclatasvir.

In the light of these circumstances, the potential clinical consequence of not reaching SVR has been strongly considered in the recommendations for treatment regimens and duration. This includes the fact

that relapse with daclatasvir is often associated with the selection of variants resistant to NS5A inhibitors. These seem to persist after discontinuation of treatment, to the extent that this has been studied, and it is still unclear whether an NS5A inhibitor would contribute to the efficacy of a retreatment regimen. Furthermore, the totality of evidence on the efficacy of sofosbuvir based DAA only regimens, and particularly sofosbuvir+NS5A inhibitor regimens, have been considered, in order to further inform tentative recommendations.

For patients with very advanced liver disease, the present attempt to reach SVR may be the last, prior to, e.g., decompensation, which may substantially impact the ultimate prognosis of the patient. It is also notable that the totality of evidence when using sofosbuvir with an NS5A inhibitor (as well as for interferon-free regimens in general), is indicative that relapse rates are somewhat lower with 24 compared to 12 weeks of therapy. It is recognised that the number needed to treat to avoid a relapse may be relatively high. This, however, has not been defined for daclatasvir+sofosbuvir. Furthermore, given the lack of specific safety concerns when using sofosbuvir+daclatasvir in combination (and the relatively low burden of side effects when ribavirin is used with this combination), there is little clinical reason for inadvertently providing a treatment regimen for cirrhotics that is not optimised in terms of the likelihood of relapse. The following regimen recommendations should be viewed in the light of these introductory comments:

Genotype 1

The AI444040 study included 126 treatment naïve, non-cirrhotic patients with genotype 1 infection. 44/44 patients treated for 24 weeks achieved SVR. 81/82 patients treated for 12 weeks achieved SVR. For one patient SVR data were missing. Needless to say, there was no impact of the addition of ribavirin on SVR. The consequent conclusion that sofosbuvir+daclatasvir without ribavirin for 12 weeks is an appropriate regimen for treatment-naïve non-cirrhotic patients is supported by other studies in which sofosbuvir is used with an NS5A inhibitor. Moreover it is noted that, while the contribution of daclatasvir or another NS5A inhibitor to a retreatment regimen in a patient who has selected for high level NS5A resistance (likely to persist, based on available data) is unknown, such previously untreated patients without cirrhosis are likely to have effective retreatment regimens available in case of relapse after the discontinuation of treatment.

As stated above, it has been demonstrated that prior exposure to peginterferon+ribavirin does not impact viral dynamics in a second treatment course. Therefore, peginterferon+ribavirin experienced patients with genotype 1 infection are considered similar to the subsection of treatment naïve patients that are most difficult to cure with this treatment modality. By the same line of argument, those patients that have failed an interferon-based triple regimen including a NS3/4A inhibitor, may be considered a further enriched subpopulation of difficult to treat patients, insofar as there is no cross-resistance between NS3/4A inhibitors and NS5A or NS5B inhibitors.

The AI444040 study contained 41 patients that had previously failed NS3/4A based triple therapy. These patients received 24 weeks of therapy with or without ribavirin. All of these patients reached SVR. There are no data in patients that previously failed on peginterferon+ribavirin alone. Furthermore, as stated above, it is not possible to tease out the individual role of the host and viral factors that have previously been associated with lower treatment response or the need for a longer treatment duration in order to maximise SVR, as all patients for whom outcome data are available reached SVR. Therefore, while it is recognised that prior treatment experience per se is not likely to impact response to sofosbuvir+daclatasvir, such experience is understood as a predefined proxy for the impact of the sum of factors with a negative impact on antiviral response.

Weighing the totality of evidence, including the fact that a treatment experienced population is functionally represented in a treatment naïve population provided that there is no cross-resistance

between drugs, it seems likely that a 12 week course of sofosbuvir+daclatasvir will give very high SVR rates also in patients preselected by prior non-response to therapy. Further, in non-cirrhotic patients previously not exposed to a DAA, effective retreatment regimens will be available. However, it is recognised that data are only available on 24 weeks of therapy with sofosbuvir+daclatasvir in treatment experienced patients. For those that have prior experience of NS3/4A inhibitors, it is presently not entirely clear when an effective retreatment regimen will be available. Therefore, it is recommended to consider adding ribavirin or prolonging therapy up to 24 weeks in such patients.

As stated in the introduction to this discussion, patients defined as cirrhotic were not included in the AI444040 study. Studies of sofosbuvir in combination with an NS5A inhibitor have indicated a higher frequency of relapse after 12 weeks of therapy in cirrhotics that also have other negative prognostic factors (which is often the case). Given the lack of specified safety concerns with sofosbuvir+daclatasvir, the negative impact of an unnecessary relapse after an insufficient duration of treatment weighs heavily as the clinical basis of a recommendation of 24 weeks of therapy in the general case in patients with cirrhosis.

Based on the totality of evidence with the use of sofosbuvir in combination with an NS5A inhibitor, a reduction of treatment duration to 12 weeks may be considered in cirrhotic patients that otherwise have positive prognostic factors such as no previous treatment failure, IL28B C/C genotype and low baseline viral load. The data to support this suggestion, however, is not generated with daclatasvir. Still, based on PK/PD considerations, they are considered likely relevant to this drug.

For patients with very advanced liver disease, including thrombocytopenia, data on the required treatment duration for maximal likelihood of SVR with interferon-free regimens are generally scarce. In such patients, the present treatment course may possibly be the last prior to decompensation, death or liver transplantation. Relapse in such patients must, if possible, be avoided. Available data indicate that such very advanced patients may generally require more drug pressure to achieve SVR. It is recognised that the contributory role of ribavirin in addition to sofosbuvir + an NS5A inhibitor is not clear in such patients. However in such cases, taking all these factors into account, the clinician may consider adding ribavirin to sofosbuvir+daclatasvir in a 24 week treatment course.

Genotype 2

Moving to genotype 2, we are left with many uncertainties regarding the most effective way to use DCV in these genotypes and how to tailor regimens according to important factors potentially effecting response. The clinical experience of sofosbuvir+daclatasvir in genotype 2 is limited to 26 patients, all of whom received 24 weeks of therapy; 9 of whom also got ribavirin. 25/26 patients achieved SVR, with data missing for one patient. Based on cross-study comparison, it is anticipated that many, perhaps most, of these patients would have achieved SVR with the background regimen alone. Furthermore, the possibility of bridging antiviral efficacy from genotype 1 is hampered by the fact that the L31M polymorphism is present in 60% of genotype 2 samples. This confers a 440-2600 fold increase in replicon EC_{50}, depending on the genotype background.

It is recognised, however, that the applicant has provided data indicative that daclatasvir contributes to regimen efficacy also in such cases. Still, it is not considered possible to define the appropriate role of daclatasvir within a treatment regimen for genotype 2 and there are no data to support the assertion that sofosbuvir+daclatasvir for 12 weeks is equally effective as sofosbuvir+ribavirin, however plausible this may seem. Furthermore, the available interferon-free treatment option is anticipated to provide SVR for near 100% of patients with genotype 2. In those few that might fail, a retreatment course of with the same drugs for a longer time is anticipated to have a high efficacy. Therefore, no regimen recommendation for daclatasvir in genotype 2 is made, available data on in vitro susceptibility and clinical experience being described in section 5.1. of the SmPC.

Genotype 3

The clinical experience of sofosbuvir+daclatasvir in genotype 3 is similarly limited as in genotype 2. A total of 18 patients have been treated for 24 weeks, five of whom also received ribavirin. 16/18 patients achieved SVR, with one confirmed relapse and one patient classified as a viral breakthrough based on an overly-strict early definition. Furthermore, it is noted that the replicon EC_{50} for genotype 3 is 43-86-fold higher when using GT1a or -1b as reference. Nonetheless, similar to the case with genotype 2, the company has presented viral kinetic data that are indicative that daclatasvir contributes to regimen efficacy in genotype 3. This conclusion is supported by outcomes in the phase II AI444031 study, where daclatasvir was used in combination with peginterferon+ribavirin. External support for this conclusion may also be derived from data for sofosbuvir in combination with another NS5A inhibitor.

In contrast to the case with genotype 2, the background regimen of only sofosbuvir+ribavirin for 24 weeks, as presently licensed, is anticipated to have a relatively high relapse rate in patients with multiple negative prognostic factors, in particular cirrhosis and prior treatment experience. In such patients, particularly if considered unsuitable for peginterferon therapy, it would be reasonable to add a further DAA that will augment the sum antiviral efficacy of the regimen. It is recognised that there is no metric on the incremental efficacy provided by adding daclatsvir to sofosbuvir+ribavirin in such cases. However, based on available data, there is a sufficient basis to consider that efficacy will be increased. Furthermore, there are no safety concerns to offset this anticipated benefit of unknown magnitude. Furthermore, it is recommended that the addition of daclatasvir to sofosbuvir+ribavirin for 24 weeks may be used in patients with negative prognostic factors such as cirrhosis and/or prior treatment experience. It is noted that there are no data to inform on the tradeoff of adding daclatasvir to the regimen in such patients, and of shortening the regimen to 12 weeks. Therefore, no shortening of therapy can be recommended.

Genotype 4

There are no data on the use of sofosbuvir+daclatasvir in genotype 4. However, there are data on the use of daclatasvir in combination with peginterferon and ribavirin in genotype 4. These are indicative that the efficacy of daclatasvir against this genotype is not lower than against genotype 1a. The in vitro potency of daclatasvir against genotype 4 in the replicon system is similar to that in genotype 1b. Furthermore, while the genetic diversity of genotype 4 is recognised, substitutions at the positions recognised to impact daclatasvir potency tend to produce lower FCs in a genotype 4 background, compared to genotype 1 (particularly genotype 1a). In summary these data are indicative that daclatasvir is as effective in genotype 4 as in genotype 1; altogether genotype 4 may be comparable to genotype 1b in terms of daclatasvir response.

It has previously been recognised that sofosbuvir efficacy is roughly similar in genotypes 4 and 1. Furthermore, genotype 4 is not intrinsically more difficult to treat than is genotype 1. Therfore, as combination effects of direct acting antiviral drugs are not anticipated to be genotype-specific, the findings in AI444040 may be extrapolated to genotype 4. Such an extrapolation has previously been accepted by the CHMP in an analogous case. Safety is anticipated to be similar regardless of genotype. In the absence of precise efficacy estimates, the sofosbuvir+daclatasvir treatment durations recommended for genotype 1 are considered relevant also for genotype 4.

Furthermore, the applicant has requested that the use of daclatsvir with pegIFN/RBV, as used in the AI444042 study, be cited as a recommended regimen in section 4.2. of the SmPC. While the, relatively speaking, inferior safety profile of interferon-based regimens is recognised, the efficacy data from this study, along with the totality of the safety database for daclatasvir when used with pegIFN/RBV, is supportive of this proposal.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considers by consensus that the risk-benefit balance of Daklinza in combination with other medicinal products in the treatment of chronic hepatitis C infection in adults is favourable and therefore recommends the granting of the marketing authorisation subject to the following conditions:

Conditions or restrictions regarding supply and use

Medicinal products on "restricted" medical prescription, reserved for use in certain specialised areas (see Annex I: Summary of Product Characteristics, section 4.2).

Conditions and requirements of the Marketing Authorisation

- **Periodic Safety Update Reports**

The marketing authorisation holder shall submit the first periodic safety update report for this product within 6 months following authorisation. Subsequently, the marketing authorisation holder shall submit periodic safety update reports for this product in accordance with the requirements set out in the list of Union reference dates (EURD list) provided for under Article 107c(7) of Directive 2001/83/EC and published on the European medicines web-portal.

Conditions or restrictions with regard to the safe and effective use of the medicinal product

- **Risk Management Plan (RMP)**

The MAH shall perform the required pharmacovigilance activities and interventions detailed in the agreed RMP presented in Module 1.8.2 of the Marketing Authorisation and any agreed subsequent updates of the RMP.

An updated RMP should be submitted:

- At the request of the European Medicines Agency;
- Whenever the risk management system is modified, especially as the result of new information being received that may lead to a significant change to the benefit/risk profile or as the result of an important (pharmacovigilance or risk minimisation) milestone being reached.

If the dates for submission of a PSUR and the update of a RMP coincide, they can be submitted at the same time.

New Active Substance Status

Based on the CHMP review of data on the quality properties of the active substance, the CHMP considers that daclatasvir is qualified as a new active substance.

APPENDIX 1: SUMMARIES EFFICACY RESULTS FOR INDIVIDUAL STUDIES

AI444040		
• **Title:** Parallel, Open-Label, Randomized Study to Evaluate the Safety, Pharmacokinetics, and Pharmacodynamics of PSI-7977 in Combination with BMS-790052 with or without Ribavirin in Treatment Naive Subjects Chronically Infected with Hepatitis C Virus Genotypes 1, 2, or 3		
Study identifier	AI444040	
Design	Randomized, open-label, pivotal study of DCV/SOF +/-RBV in treatment-naïve subjects with GT-1, -2, and -3 and in GT-1 subjects who had previously failed a TVR/BOC based regimen. Subjects with baseline cirrhosis were excluded. There were 10 treatment groups:	
	<u>24 weeks of treatment</u> Group A: SOF 400 mg QD x 7 days then add DCV 60 mg QD Group B: SOF 400 mg QD x 7 days then add DCV 60 mg QD Group C: DCV 60 mg QD + SOF 400 mg QD Group D: DCV 60 mg QD + SOF 400 mg QD Group E: DCV 60 mg QD + SOF 400 mg QD + RBV Group F: DCV 60 mg QD + SOF 400 mg QD + RBV	 15 GT-1a/1b 16 GT-2/-3 14 GT-1a/-1b 14 GT-2/-3[a] 15 GT-1a/1b 14 GT-2/-3
	<u>12 weeks of treatment</u> Group G: DCV 60 mg QD + SOF 400 mg QD Group H: DCV 60 mg QD + SOF 400 mg QD + RBV	 41 GT-1a/1b 41 GT-1a/1b
	<u>24 weeks of treatment</u> Group I: DCV 60 mg QD + SOF 400 mg QD Group J: DCV 60 mg QD + SOF 400 mg QD + RBV	 21 GT-1a/1b 20 GT-1a/1b
	Duration of main phase	12 or 24 weeks as described above
	Duration of Follow-up phase	48 weeks follow-up for all groups
	Rescue therapy	Subjects who had virologic failure on treatment could have added pegIFN □ not receiving RBV as part of their original treatment regimen) or pegIFN □ a subjects receiving RBV as part of their original treatment regimen) to be their DAA regimen.
Hypothesis	A combination of SOF and DCV will be identified with or without RBV, which provides potent antiviral activity and prevents emergence of resistance in multiple HCV genotypes.	
Treatment groups	See the design section above.	

AI444040			
Endpoints and definitions	Primary endpoint	Primary endpoint	Proportion of subjects with SVR12, defined as HCV RNA below LLOQ (< 25 IU/mL), target detected (TD) or target not detected (TND) at follow-up Week 12
	Secondary endpoint	Secondary endpoint(s)	a) Proportion of subjects who achieved HCV RNA < LLOQ, TD or TND at Weeks 1, 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, and 22 weeks of therapy; at end of treatment (EOT, following 12 or 24 weeks of treatment, by group); and follow-up Weeks 4, 12, 24, 36, and 48 b) Proportion of subjects who achieved HCV RNA < LLOQ, TND at Weeks: 1, 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, and 22 weeks of therapy; at EOT (following 12 or 24 weeks of treatment by group); and follow-up Weeks 4, 12, 24, 36, and 48 c) To describe rates of viral breakthrough (VBT) and relapse d) To characterize the development of antiviral resistance through HCV genomic substitutions e) To estimate the rate of sustained virologic response at follow-up Week 24 (SVR24) defined as HCV RNA < LLOQ, TD or TND at follow-up Week 24
Database lock	18-Nov-2013		

Results and Analysis

Analysis description	**Primary Analysis - Sustained Virologic Response at Follow-up Week 12**			
Analysis population and time point description	Primary efficacy population was based on the modified intent-to-treat (mITT) population where the numerator was based on subjects who met the response criteria (at follow-up Week 12). The denominator was based on all treated subjects.			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/- RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/-RBV	TVR/BOC Failures GT-1 DCV/SOF +/- RBV
	Number of subjects	126	44	41
	SVR12 (Responder; %)	124 (98.4)	40 (90.9)	40 (97.6)
	SVR12 with imputation**	125 (99.2)	41 (93.2) GT-2 25/26 (96.2) GT-3 16/18 (88.9)	41 (100.0)
Notes	*Defined as HCV RNA below LLOQ (< 25 IU/mL) TD or TND at follow-up Week 12 **Subjects with missing HCV RNA at follow-up Week 12 were counted as SVR12 responders if they had HCV RNA < LLOQ, TD or TND at the next available measurement			

AI444040				
Analysis description	**Secondary analysis - Week 4 Virologic Response: HCV RNA < LLOQ, TD or TND at Week: 4**			
Analysis population and time point description	Secondary analyses were based on the modified intent-to-treat (mITT) population where the numerator was based on subjects who met the response criteria. The denominator was based on all treated subjects at visit weeks defining the endpoint.			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF+/-RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/-RBV	TVR/BOC Failures GT-1 DCV/SOF+/-RBV
	Number of subjects	126	44	41
	(Responder; %) at Week 4	124 (98.4)	44 (100.0)	40 (97.6)
Analysis description	**Secondary analysis - EOTR defined as HCV RNA < LLOQ, TD or TND at EOT**			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/- RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/- RBV	TVR/BOC Failures GT-1 DCV/SOF +/-RBV
	Number of subjects	126	44	41
	HCV RNA < LLOQ, TD or TND at EOT (Responder; %)	126 (100.0)	43 (97.7)	41 (100.0)
Analysis description	**Secondary analysis - SVR24: HCV RNA < LLOQ, TD or TND at follow-up Week 24**			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/- RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/- RBV	TVR/BOC Failures GT-1 DCV/SOF +/-α RBV
	Number of subjects	126	44	41
	SVR24 (Responder; %)	120 (95.2)	41 (93.2)	41 (100.0)
Analysis description	**Secondary analysis -Rapid virologic response (RVR) HCV RNA < LLOQ, TND at Week 4**			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/-α RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/- RBV	TVR/BOC Failures GT-1 DCV/SOF +/-α RBV
	Number of subjects	126	44	41
	RVR (Responder; %)	100 (79.4)	34 (77.3)	31 (75.6)

AI444040				
Analysis description	**Secondary analysis - EOTR defined as HCV RNA < LLOQ, TND at EOT**			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/-RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/- RBV	TVR/BOC Failures GT-1 DCV/SOF +/- RBV
	Number of subjects	126	44	41
	HCV RNA < LLOQ, TND at EOT (Responder; %)	126 (100.0)	42 (95.5)	38 (92.7)
Analysis description	**Secondary analysis - VBT and Relapse through follow-up Week 12/24**			
Descriptive statistics	Treatment group	Treatment-naive Subjects with GT-1 DCV/SOF +/- RBV	Treatment-naive Subjects with GT-2/-3 DCV/SOF +/- RBV	TVR/BOC Failures GT-1 DCV/SOF +/- RBV
	Number of subjects	126	44	41
	VBT*	0	1 (2.3)	0
	Relapse**	1 (0.8)	1 (2.3)	0
Notes	* VBT defined as: **Original Protocol Definition of VBT** 5. Any increase in HCV viral load ≥ 1 log from nadir (not necessarily from a consecutive sampling). 6. Any confirmed HCV RNA < LLOQ, TD on or after Week 8 (i.e., 2 consecutive results of HCV RNA < LLOQ, TD). 7. Any HCV RNA < LLOQ on or after Week 8 (no confirmation needed). **Protocol Amendment 03/05 Definition of VBT** 1. Any confirmed increase in viral load ≥ 1 log from nadir (included in Protocol Amendment 03, but the word confirmed was added in Protocol Amendment 05) 2. Any confirmed HCV RNA ≥ 25 IU/mL (e.g., HCV RNA > limit of quantitation) on or after Week 8 (included in Protocol Amendment 03) **Definition of viral relapse:** HCV RNA>LLOQ during follow-up after HCV RNA < LLOQ, TD or TND at EOT.			

AI444040	
Analysis description	**Secondary analysis - To characterize the development of antiviral resistance through HCV genomic substitutions**
Descriptive statistics	• Baseline NS5A resistance-associated polymorphisms at amino acid positions 28, 30, 31, and/or 93 that have been shown to confer loss in DCV potency in vitro were observed in 33/203 (16.3%) subjects. o The most common NS5A resistance-associated polymorphisms, L31M and Y93H/N/C, were detected at baseline in 8/203 (3.9%) and 20/203 (9.9%) of subjects, respectively. • No baseline NS5B resistance-associated polymorphisms at S282T were detected. • There did not appear to be a relationship with baseline NS5A resistance-associated polymorphisms and virologic response. o All subjects with pre-existing DCV resistance variants achieved SVR, with the exception of 1 GT-3 subject. This subject had an NS5A-A30K polymorphism at baseline and at relapse.

BOC - boceprevir, DCV - daclatasvir, EOT - end of treatment, EOTR - end of treatment response, GT - genotype, HCV - hepatitis C virus, ITT - intent-to-treat, LLOQ - lower limit of quantitation, RNA - ribonucleic acid, RBV - ribavirin, SOF - sofosbuvir, SVR12, 24 - sustained virologic response (HCV RNA < LLOQ, TD or TND) at follow-up Weeks 12 and 24, TD - target detected, TND - target not detected, TVR - telaprevir, VBT - virologic breakthrough

AI444010	
• **Title:** A Phase 2b Study of BMS-790052 in Combination with Peg-Interferon Alfa-2a and Ribavirin in Treatment-naïve Subjects with Chronic Hepatitis C Genotype 1 and 4 Infection	
Study identifier	AI444010
Design	Randomized, double-blind, placebo-controlled Phase 2b study: **Stage 1:** All treatment-naïve GT-1 and GT-4 HCV-infected subjects (randomized 2:2:1) received DCV/pegIFNα/RBV or placebo/pegIFNα/RBV through Week 12. **Stage 2:** At Week 12, a second randomization (1:1) occurred for subjects initially randomized to DCV/pegIFNα/RBV who achieved a protocol-defined response (PDR: HCV RNA < LLOQ, target detected [TD] or TND at Week 4 and HCV RNA < LLOQ, TND at Week 10), these subjects received an additional 12 weeks of DCV/pegIFNα/RBV or 12 weeks of placebo/pegIFNα/RBV. At Week 12, subjects initially randomized to DCV/pegIFNα/RBV who did not achieve PDR received an additional 36 weeks of therapy: 12 weeks of placebo/pegIFNα/RBV followed by 24 weeks of pegIFNα/RBV. All subjects initially randomized to placebo (regardless of PDR status) received an additional 36 weeks of therapy: 12 weeks placebo/pegIFNα/RBV followed by 24 weeks of pegIFNα/RBV.
Duration of main phase (Stage 1 and Stage 2)	Up to 24 or 48 weeks on-treatment: f) Double-blind DCV/pegIFNα/RBV or placebo/pegIFNα/RBV up to 24 weeks for subjects initially randomized to DCV/pegIFNα/RBV who achieved PDR (12 weeks DCV/pegIFNα/RBV + 12 weeks DCV/pegIFNα/RBV or 12 weeks DCV/pegIFN+/-RBV + 12 weeks placebo/pegIFNα/RBV) g) Therapy with pegIFNα/RBV for up to an additional 24 weeks for 1) all subjects initially randomized to placebo/pegIFNα/RBV regardless of PDR status, and 2) subjects initially randomized to DCV/pegIFNα/RBV who did not achieve PDR
Duration of follow-up	Up to 24 or 48 weeks
Hypothesis	At least 1 dose of DCV combined with pegIFN+/-RBV can be identified which is safe, well tolerated, and demonstrates eRVR rates 35% greater than control (placebo/pegIFN+/-RBV) in treatment-naïve chronically-infected HCV GT-1 subjects. At least 1 dose of DCV combined with pegIFN+/-RBV can be identified which is safe, well tolerated, and demonstrates SVR rates which are superior to control (placebo/pegIFNR+/-BV) in treatment-naïve, chronically-infected HCV GT-1 subjects.
Treatment groups	395 subjects were randomized 2:2:1 (DCV 20 mg:DCV 60 mg:placebo) 365 subjects with HCV GT-1: 147 treated with DCV 20 mg/pegIFN+/-/RBV, 146 treated with DCV 60 mg/ pegIFNα/RBV, and 72 treated with placebo/ pegIFN+/-/RBV 30 subjects with GT-4: 12 treated with DCV 20 mg/pegIFN+/-/RBV, 12 treated with DCV 60 mg/ pegIFN+/-/RBV, and 6 treated with placebo/ pegIFN+/-/RBV

AI444010			
Endpoints and definitions	Primary endpoints	Primary endpoint	Proportion of subjects with eRVR defined as HCV RNA < LLOQ (25 IU/mL), TND at both Weeks 4 and 12
		Co-primary endpoint	Proportion of subjects with SVR24 defined as HCV RNA < LLOQ, TND at follow-up Week 24
	Secondary endpoints	Key Secondary endpoints	Proportion of subjects with RVR defined as HCV RNA < LLOQ, TND at Week 4 on-treatment Proportion of subjects with cEVR defined as HCV RNA < LLOQ, TND at Week 12 on-treatment Proportion of subjects with SVR12 defined as HCV RNA < LLOQ, TND at follow-up Week 12 Frequency of genotypic substitutions at baseline, on-treatment and during follow-up associated with DCV virologic failure
Database lock	16-Nov-2012		

Results and Analysis

Analysis description	**Primary Analysis - Extended Rapid Virologic Response**
Analysis population and time point description	Antiviral activity endpoints were summarized by treatment regimen using modified ITT (mITT): the numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects. Response rates and 80% exact binomial CIs were presented by treatment group using mITT and observed values. CIs are based on the normal approximation to the binomial distribution.

Descriptive statistics and estimate variability	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFN+/- / RBV	DCV 60 mg + pegIFN+/-/ RBV	Placebo + pegIFN+/-/ RBV	DCV 20 mg + pegIFN+/- / RBV	DCV 60 mg + pegIFN+/-/ RBV	Placebo + pegIFN+/-/ RBV
	Number of subjects	147	146	72	12	12	6
	eRVR* Responder (%)	80 (54.4)	79 (54.1)	10 (13.9)	2 (16.7)	4 (33.3)	0 (0)
	80% CI	(49.2, 59.7)	(48.8, 59.4)	(8.7, 19.1)	(2.9, 30.5)	(15.9, 50.8)	(0.0, 0.0)
	Difference DCV - Placebo (80% CIs)	40.5 (33.1, 47.9)	40.2 (32.8, 47.7)	-	16.7 (2.9, 30.5)	33.3 (15.9, 50.8)	-
Notes	* Defined as HCV RNA < LLOQ, TND at both Weeks 4 and 12						

AI444010							
Analysis description	**Primary Analysis - Sustained Virologic Response at Follow-up Week 24**						
Descriptive statistics and estimate variability	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFN+/- RBV	DCV 60 mg + pegIFN+/ -RBV	Placebo + pegIFN+ /- RBV	DCV 20 mg + pegIFN+ /- RBV	DCV 60 mg + pegIFN +/-RBV	Placebo + pegIFNR +/- BV
	Number of subjects	147	146	72	12	12	6
	SVR24* Responder (%)	87 (59.2)	87 (59.6)	27 (37.5)	8 (66.7)	12 (100.0)	3 (50.0)
	80% CI	(54.0, 64.4)	(54.4, 64.8)	(30.2, 44.8)	(49.2, 84.1)	(100.0, 100.0)	(23.8, 76.2)
Notes	* Defined as HCV RNA < LLOQ, TND at follow-up Week 24						
Analysis description	**Secondary Analysis - Rapid Virologic Response**						
Descriptive statistics and estimate variability	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFNR+ /-BV	DCV 60 mg + pegIFN +/-RBV	Placebo + pegIFN+ /-RBV	DCV 20 mg + pegIFNR +/-BV	DCV 60 mg + pegIFN RBV+/-	Placebo + pegIFN+ /- RBV
	Number of subjects	147	146	72	12	12	6
	RVR* Responder (%)	88 (59.9)	83 (56.8)	11 (15.3)	3 (25.0)	4 (33.3)	0 (0)
	80% CI	(54.7, 65.0)	(51.6, 62.1)	(51.6, 62.1)	(9.0, 41.0)	(15.9, 50.8)	(0.0, 0.0)
Notes	* Defined as HCV RNA < LLOQ, TND at Week 4 on-treatment						
Analysis description	**Secondary Analysis - Complete Early Virologic Response**						
Descriptive statistics and estimate variability	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFN +/-RBV	DCV 60 mg + pegIFN+/ - RBV	Placebo + pegIFN+ /- RBV	DCV 20 mg + pegIFNR +/-BV	DCV 60 mg + pegIFN +/-RBV	Placebo + pegIFN+ /- RBV
	Number of subjects	147	146	72	12	12	6
	cEVR* Responder (%)	114 (77.6)	110 (75.3)	31 (43.1)	9 (75.0)	12 (100.0)	3 (50.0)
	80% CI	(73.1,	(70.8,	(35.6,	(59.0,	(100.0,	(23.8,

	82.0)	79.9)	50.5)	91.0)	100.0)	76.2)

Notes	* Defined as HCV RNA < LLOQ, TND at Week 12 on-treatment
Analysis description	**Secondary Analysis - Sustained Virologic Response at Follow-up Week 12**

Descriptive statistics and estimate variability	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFN+/-RBV	DCV 60 mg + pegIFN +/-RBV	Placebo + pegIFNR+ /-BV	DCV 20 mg + pegIFN +/-RBV	DCV 60 mg + pegIFN+/-RBV	Placebo + pegIFN +/-RBV
	Number of subjects	147	146	72	12	12	6
	SVR12* Responder (%)	95 (64.6)	88 (60.3)	26 (36.1)	9 (75.0)	12 (100.0)	3 (50.0)
	80% CI	(59.6, 69.7)	(55.1, 65.5)	(28.9, 43.4)	(59.0, 91.0)	(100.0, 100.0)	(23.8, 76.2)
Notes	* Defined as HCV RNA < LLOQ, TND at follow-up Week 12						

Analysis description	**Secondary Analysis - Virologic Failure**

	Treatment group	GT-1			GT-4		
		DCV 20 mg + pegIFN +/-RBV	DCV 60 mg + pegIFN+/-RBV	Placebo + pegIFN+/- RBV	DCV 20 mg + pegIFN +/-RBV	DCV 60 mg + pegIFN +/-RBV	Placebo + pegIFNR +/-BV
	Number of subjects	147	146	72	12	12	6
	VBT	8.2 (12/147)	10.3 (15/146)	2.8 (2/72)	8.3 (1/12)	0	0
	Relapse	18.5 (22/119)	19.0 (22/116)	22.0 (9/41)	20.0 (2/10)	0 (0/12)	25.0 (1/4)

Virologic failure, for the purpose of the study, was defined as:

- VBT: confirmed > 1 log_{10} increase in HCV RNA over nadir or confirmed HCV RNA □ LLOQ after confirmed HCV RNA < LLOQ, TND while on treatment. Measurements were confirmed at the next scheduled visit.
- < 1 log_{10} decrease in HCV RNA from baseline at Week 4 of treatment
- Failure to achieve early virologic response (EVR): < 2 log_{10} decrease in HCV RNA from baseline and HCV HCV RNA ≥ LLOQ at Week 12 of treatment
- HCV RNA < LLOQ, TD or ≥ LLOQ at Week 12 and ≥ LLOQ at Week 24
- HCV RNA < LLOQ, TD or ≥ LLOQ at EOT (including early discontinuation)
- Relapse, defined as HCV RNA ≥LLOQ or HCV RNA < LLOQ, TD during follow-up after HCV RNA < LLOQ, TND at EOT.

AI444010	
	A brief summary of the resistance results is provided below: • Baseline NS5A polymorphisms at L31I/V/M and Y93H/N/S in GT-1a subjects may be loosely associated with virologic failure, especially when combined with a non-CC IL-28B GT. A correlation could not be determined for baseline NS5A polymorphisms at M28 or Q30. o Any potential correlation with baseline NS5A polymorphisms at 28, 30, 31, or 93 and GT-1b and GT-4 failures was less apparent. o IL-28B GT did appear to be more predictive of failure against subjects infected with GT-1b and GT-4. • In all available subjects who failed with HCV RNA [redacted] were detected; substitutions at Q30 predominated in GT-1a, substitutions at L31-Y93 predominated in GT-1b, and substitutions at L28-L30 predominated in GT-4. • A greater number of GT-1a subjects (46%, 101/220) did not achieve SVR24 than GT-1b subjects (25%, 18/72) or GT-4 subjects (16%, 4/25). o The resistance barrier to DCV in GT-1a subjects was lower than for GT-1b and GT-4 in that one emergent substitution could confer high level resistance to DCV in GT-1a whereas at least 2 substitutions were generally required in GT-1b and GT-4. o Pre-existence of a GT-1a NS5A resistance-associated variant may increase a subject's chance of failure to DCV/pegIFN [redacted] observation is based on a limited number of cases. • Irrespective of GT or emergent variant, the emergent NS5A resistance variants were fit and generally persisted out to follow-up Week 48. • The commercially available VERSANT HCV Genotype 2.0 (LiPA) genotyping kit was shown to be reliable for GT-1 sub-typing of baseline samples from 317 subjects; mis-genotyping, as determined by NS5A sequence alignment with GT-1a (H77c) and GT-1b (Con1) reference strains, was only detected in ~ 1% of samples.

DCV - daclatasvir, GT(s) - genotype(s), EOR - end of treatment response, eRVR - extended rapid virologic response, GT - genotype, HCV - hepatitis C virus, mITT - modified intent-to-treat, PBO - placebo, PDR - protocol defined response, pegIFN - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR24 - sustained virologic response at follow-up Week 24, TD - target detected, TND - target not detected, VBT - virologic breakthrough

AI444021			
• **Title:** A Phase 2a Study of BMS-790052 in Combination with Peginterferon Alfa-2b (PegIntron®) and Ribavirin (Rebetol®) in Japanese Subjects with Genotype 1 Chronic Hepatitis C virus (HCV) Infection			
Study identifier	AI444021		
Design	Double-blind, randomized, Phase 2a study conducted in Japan where treatment-naïve subjects were administered DCV/pegIFNα/RBV or placebo/pegIFNα/RBV, and prior non-responders were administered DCV/pegIFNα/RBV		
	Duration of main phase	Double-blind DCV/pegIFNα/RBV or placebo/pegIFNα/RBV up to 24 or 48 weeks	
	Duration of follow-up phase	4 or 24 weeks of post-treatment follow-up	
Hypothesis	Based on 12-week on-treatment data, at least 1 dose of DCV can be identified which is safe, well tolerated, and efficacious when combined with pegIFNα and RBV for the treatment of chronically infected HCV GT-1 treatment-naïve and non-responder to standard of care subjects.		
Treatment groups	45 subjects (treatment- naïve and prior non-responders) were randomized 1:1:1 (treatment-naïve) and 1:1 (non-responders)		
	Placebo - Treatment-naïve	Treatment- naïve subjects were administered placebo/pegIFNα/RBV for up to 48 weeks	
	DCV 10 mg QD Treatment-naïve	Treatment-naïve subjects were administered DCV 10 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR: HCV RNA < LLOQ [15 IU/mL] at Week 4 and undetectable HCV RNA at Week 12) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 60 mg QD Treatment-naïve	Treatment-naïve subjects were administered DCV 60 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 10 mg QD Prior non-responder	Non-responder subjects were administered DCV 10 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 60 mg QD Prior non-responder	Non-responder subjects were administered DCV 60 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
Endpoints and definitions	Primary endpoint	Primary endpoint	Extended rapid virologic response (eRVR) rate defined as undetectable HCV RNA (< LLOQ, TND) at both Weeks 4 and 12 on-treatment
	Secondary endpoint	Secondary endpoint	h) Proportion of subjects with RVR i.e., HCV RNA < LLOQ (15 IU/mL), TND at Week 4 on treatment i) Proportion of subjects with cEVR, i.e., HCV RNA < LLOQ, TND at Week 12 on treatment j) Proportion of subjects with SVR12, i.e., HCV RNA < LLOQ, TND at follow-up Week 12 k) Proportion of subjects with SVR24, i.e., HCV RNA < LLOQ, TND at follow-up Week 24. l) Frequency of vial genotypic substitutions associated with virologic failure
Database lock	12-Sep-2011		
Results and Analysis			

AI444021						
Analysis description	**Primary Analysis - Extended Rapid Virologic Response**					
Analysis population and time point description	Extended rapid virologic response rates (eRVR) and exact binomial CIs were presented by treatment group using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	eRVR* Responder, (%)	0	6 (66.7)	8 (80.0)	5 (55.6)	2 (22.2)
	80% CI	(0.0, 25.0)	(40.1, 87.1)	(55.0, 94.5)	(30.1, 79.0)	(6.1, 49.0)
Notes	* On-treatment undetectable HCV RNA (< LLOQ, TND) at both Weeks 4 and 12					
Analysis description	**Secondary analysis - Rapid Virologic Response**					
Analysis population and time point description	Secondary binary efficacy endpoints (RVR, EVR, SVR, PDR, EOTR, SVR4, SVR12, and SVR24 were assessed with response rates and exact binomial CIs by treatment group using mITT.					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	RVR* Responder, (%)	0	7 (77.8)	8 (80.0)	5 (55.6)	3 (33.3)
	80% CI	(0.0, 25.0)	(51.0, 93.9)	(55.0, 94.5)	(30.1, 79.0)	(12.9, 59.9)
Notes	* On-treatment undetectable HCV RNA (< LLOQ, TND) at Week 4					

AI444021						
Analysis description	**Secondary analysis - Complete Early Virologic Response**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	cEVR* Responder, (%)	5 (62.5)	7 (77.8)	10 (100)	5 (55.6)	5 (55.6)
	80% CI	(34.5, 85.3)	(51.0, 93.9)	(79.4, 100.0)	(30.1, 79.0)	(30.1, 79.0)
Notes	* On-treatment undetectable HCV RNA (< LLOQ, TND) at Week 12					
Analysis description	**Secondary analysis - Sustained Virologic Response at Follow-up Week 12**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	SVR12* Responder, (%)	5 (62.5)	6 (66.7)	9 (90.0)	2 (22.2)	3 (33.3)
	80% CI	(34.5, 85.3)	(40.1, 87.1)	(66.3, 99.0)	(6.1, 49.0)	(12.9, 59.9)
Notes	* Undetectable HCV RNA (< LLOQ, TND) at follow-up Week 12					
Analysis description	**Secondary analysis - Sustained Virologic Response at Follow-up Week 24**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	SVR24* Responder, (%)	5 (62.5)	6 (66.7)	9 (90.0)	2 (22.2)	3 (33.3)
	80% CI	(34.5, 85.3)	(40.1, 87.1)	(66.3, 99.0)	(6.1, 49.0)	(12.9, 59.9)
Notes	* Undetectable HCV RNA (< LLOQ, TND) at follow-up Week 24					

AI444021						
Analysis description	**Secondary analysis - Virologic Failure (Treated Subjects)**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	10	9	9
	Virologic failure (%)	3 (37.5)	3 (33.3)	1 (10.0)	7 (77.8)	6 (66.7)
	VBT (%)*	1 (12.5)	1 (11.1)	0	4 (44.4)	4 (44.4)
	Relapse (%)**	2 (25.0)	1 (11.1)	1 (10.0)	3 (33.3)	2 (22.2)
	*VBT defined as confirmed > 1 log10 increase in HCV RNA over nadir or confirmed HCV RNA ≥ LOQ after confirmed undetectable HCV RNA while on treatment. Measurements were confirmed at the next scheduled assessment < 1 log10 decrease in HCV RNA from baseline at Week 4 of treatment. **Relapse, defined as detectable HCV RNA during follow-up after undetectable HCV RNA at EOT					
	m) The most predominant NS5A resistance substitutions were at amino acid residues 31(L31 changing to M or V) and 93 (Y93 changing to H). n) Information on the IL28B allele was available for 16/17 subjects who had emergent NS5A resistance-associated substitutions; 15/16 carried the non-CC allele indicating a correlation with virologic outcome. o)					

BMS - Bristol-Myers Squibb, DCV - daclatasvir, GT(s) - genotype(s), EOTR - end of treatment response, eRVR - extended rapid virologic response, EVR - early virologic response, HCV - hepatitis C virus, ITT - intent-to-treat, LOQ - limit of quantification, mITT - modified intent-to-treat, PDR -protocol defined response, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR4- sustained virologic response at follow-up Week 4, SVR12 - sustained virologic response at follow-up Week 12, SVR24 - sustained virologic response at follow-up Week 24, VBT - virologic breakthrough

AI444022			
• **Title:** A Phase 2a Study of BMS-790052 in Combination with Peginterferon Alfa-2a (Pegasys®) and Ribavirin (Copegus®) in Japanese Subjects with Genotype 1 Chronic Hepatitis C Virus (HCV) Infection			
Study identifier	AI444022		
Design	Double-blind, randomized, Phase 2a study in treatment-naïve Japanese subjects administered DCV/ pegIFNα-2a/RBV or placebo/pegIFNα-2a/RBV, and non-responder Japanese subjects were administered DCV/pegIFNα-2a/RBV in the double-blind period for the first 24 weeks.		
	Duration of main phase	Double-blind DCV/pegIFNα/RBV or placebo/pegIFNα/RBV up to 24 or 48weeks on-treatment	
	Duration of follow-up phase	4 or 24 weeks of post-treatment follow-up	
Hypothesis	Based on 12-week on-treatment data, at least 1 dose of DCV can be identified which is safe, well tolerated, and efficacious when combined with pegIFNα/RBV for the treatment of chronically infected HCV GT-1 treatment-naïve and non-responder to standard of care subjects.		
Treatment groups	43 subjects were randomized 1:1:1 (treatment- naïve) and 1:1 (non-responders)		
	Placebo - Treatment-naïve	Treatment-naïve subjects received placebo/pegIFNα/RBV for up to 48 weeks.	
	DCV 10 mg QD - Treatment-naïve	Treatment-naïve subjects were administered DCV 10 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR: HCV RNA < LLOQ [15 IU/mL] at Week 4 and undetectable HCV RNA [< LLOQ, TND] at Week 12) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 60 mg QD - Treatment-naïve	Treatment-naïve subjects received DCV 60 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 10 mg QD - non-responder	Non-responder subjects received DCV 10 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
	DCV 60 mg QD - non-responder	Non-responder subjects received DCV 60 mg QD/pegIFNα/RBV for up to 24 weeks (subjects who achieved PDR) or for up to 48 weeks (subjects who did not achieve eRVR).	
Endpoints and definitions	Primary endpoint	Primary endpoint	Extended rapid virologic response (eRVR) rate defined as undetectable HCV RNA (< LLOQ, TND) at both Weeks 4 and 12 on-treatment
	Secondary endpoints	Secondary endpoints	Proportion of subjects with RVR, defined as undetectable HCV RNA (< LLOQ, TND) at Week 4 on-treatment Proportion of subjects with cEVR, defined as undetectable HCV RNA (< LLOQ, TND) at Week 12 on-treatment Proportion of subjects with SVR12, defined as undetectable HCV RNA (< LLOQ, TND) at follow-up Week 12 Proportion of subjects with SVR24, defined as undetectable HCV RNA (< LLOQ, TND) at follow-up Week 24 Resistant variants associated with virologic failure
Database lock	29-Nov-2011		

AI444022						
Results and Analysis						
Analysis description	**Primary Analysis - Extended Rapid Virologic Response**					
Analysis population and time point description	Extended rapid virologic response rates (eRVR) and exact binomial CIs were presented by treatment group using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects					
Descriptive statistics and estimate variability	Treatment group	Treatment- naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	eRVR* Responder, (%)	1 (12.5)	6 (66.7)	5 (62.5)	5 (62.5)	7 (77.8)
	80 % CIs	(1.3, 40.6)	(40.1, 87.1)	(34.5, 85.3)	(34.5, 85.3)	(51.0, 93.9)
Notes	* On-treatment undetectable HCV RNA at both Weeks 4 and 12					
Analysis description	**Secondary analysis - Rapid Virologic Response**					
Analysis population and time point description	Secondary binary efficacy endpoints (RVR, cEVR, EVR, PDR, EOTR, SVR4, SVR12, SVR24, undetectable RNA and HCV RNA AUCO over time, and HCV RNA changes from baseline) were assessed with response rates and exact binomial CIs by treatment group using the modified ITT.					
Descriptive statistics and estimate variability	Treatment group	Treatment naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	RVR* Responder, (%)	1 (12.5)	7 (77.8)	5 (62.5)	5 (62.5)	8 (88.9)
	80% CI	(1.3, 40.6)	(51.0, 93.9)	(34.5, 85.3)	(34.5, 85.3)	(63.2, 98.8)
Notes	* On-treatment undetectable HCV RNA at Week 4					
Analysis description	**Secondary analysis - Complete Early Virologic Response**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	cEVR* Responder, (%)	5 (62.5)	8 (88.9)	8 (100.0)	7 (87.5)	8 (88.9)
	80% CI	(34.5, 85.3)	(63.2, 98.8)	(75.0, 100.0)	(59.4, 98.7)	(63.2, 98.8)
Notes	* On-treatment undetectable HCV RNA at Week 12					

AI444022						
Analysis description	**Secondary analysis - Sustained Virologic Response at Follow-up Week 12**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	SVR12* Responder, (%)	6 (75.0)	8 (88.9)	8 (100.0)	4 (50.0)	7 (77.8)
	80% CI	(46.2, 93.1)	(63.2, 98.8)	(75.0, 100.0)	(24.0, 76.0)	(51.0, 93.9)
Notes	* Undetectable HCV RNA at follow-up Week 12					
Analysis description	**Secondary analysis - Sustained Virologic Response at Follow-up Week 24**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	SVR24* Responder, (%)	6 (75.0)	8 (88.9)	8 (100.0)	4 (50.0)	7 (77.8)
	80% CI	(46.2, 93.1)	(63.2, 98.8)	(75.0, 100.0)	(24.0, 76.0)	(51.0, 93.9)
Notes	* Undetectable HCV RNA at follow-up Week 24					
Analysis description	**Secondary analysis - Virologic Failure (Treated Subjects)**					
Descriptive statistics and estimate variability	Treatment group	Treatment-naïve			Non-responder	
		Placebo	DCV 10 mg	DCV 60 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	8	9	8	8	9
	Virologic failure (%)	1 (12.5)	1 (11.1)	0	4 (50.0)	2 (22.2)
	VBT (%)*	0	1 (11.1)	0	1 (12.5)	1 (11.1)
	Relapse (%)**	1 (12.5)	0	0	3 (37.5)	1 (11.1)
Notes	* VBT defined as confirmed > 1 $\log_{10}$ increase in HCV RNA over nadir or confirmed HCV RNA treatment. **Relapse, defined as detectable HCV RNA during follow-up after undetectable HCV RNA at EOT					

AI444022	
	p) For all 7 DCV-treated subjects experiencing virologic failure, emergent NS5A resistance-associated substitutions were detected. q) The most predominant NS5A resistance substitutions were at amino acid residues 31(L31 changing to M or V) and 93 (Y93 changing to H). r) Information on the IL28B allele was available for all 7 subjects who had emergent NS5A resistance-associated substitutions; 7/7 carried the non-CC allele indicating a correlation with virologic outcome. s) A correlation between pre-existing NS5A resistance-associated polymorphisms and virologic outcome could not be determined in this small study; 9 subjects with resistance-associated substitutions at L28M, Q30R, R30Q, P58L/S, and/or Y93H responded while 2 subjects with L28M, R30Q, and/or P58S experienced virologic failure.

BMS - Bristol-Myers Squibb, cEVR - complete early virologic response, CI(s) - confidence interval(s), DCV - daclatasvir, GT(s) - genotype(s), EOTR - end of treatment response, eRVR - extended rapid virologic response, HCV - hepatitis C virus, LLOQ - less than the limit of quantitation, mITT - modified intent-to-treat, PDR -protocol defined response, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR12, 24 - sustained virologic response at follow-up Weeks 12 and 24, respectively, TND - target not detected, VBT - virologic breakthrough

AI444014			
• **Title: A Phase 2a Study of BMS-790052 in Combination with Peginterferon Alfa-2a (Pegasys®) and Ribavirin (Copegus®) in Treatment-Naïve Subjects with Chronic Hepatitis C Virus Genotype 1 Infection**			
Study identifier	AI444014		
Design	Randomized, double-blind, placebo-controlled Phase 2a study in which treatment-naïve GT-1 HCV-infected subjects were administered DCV/pegIFNα/RBV or placebo/pegIFNα/RBV		
	Duration of main phase	Double-blind DCV/pegIFNα/RBV or placebo/pegIFNα/RBV up to 48weeks on-treatment	
	Duration of follow-up phase	24 weeks of post-treatment follow-up	
Hypothesis	Based on 12-week on-treatment data, at least 1 dose of DCV can be identified which is safe, well tolerated, and efficacious when combined with pegIFN of chronically infected HCV GT-1 treatment-naïve subjects.		
Treatment groups	48 treatment- naïve subjects were randomized (1:1:1:1)		
	Placebo - naïve		Treatment-naïve subjects received placebo/pegIFNα/RBV for up to 48 weeks.
	DCV 3 mg QD - naïve		Treatment-naïve subjects received DCV 3 mg QD/pegIFNα/RBV for up to 48 weeks
	DCV 10 mg QD - naïve		Treatment-naïve subjects received DCV 10 mg QD/pegIFNα/RBV for up to 48 weeks
	DCV 60 mg QD - naïve		Treatment-naïve subjects received DCV 60 mg QD/pegIFNα/RBV for up to 48 weeks
Endpoints and definitions	Primary endpoint	Primary endpoint	Extended rapid virologic response (eRVR) rate defined as undetectable HCV RNA (< LLOQ, TND) at both Weeks 4 and 12 on-treatment
	Secondary endpoints	Secondary endpoints	t) Proportion of subjects with RVR, defined as undetectable HCV RNA (< LLOQ, TND)at Week 4 on-treatment u) Proportion of subjects with EVR, defined as ≥ 2 $\log_{10}$ decrease in HCV RNA from baseline at Week 12 or HCV RNA < 10 IU/mL on-treatment for subjects with baseline HCV RNA < 1000 IU/mL v) Proportion of subjects with SVR12, defined as undetectable HCV RNA (< LLOQ, TND)at follow-up Week 12 w) Proportion of subjects with SVR24, defined as undetectable HCV RNA (< LLOQ, TND)at follow-up Week 24 x) Resistant variants associated with clinical failure
Database lock	Final CSR (SVR12): 08-Dec-2010 Addendum 01 (SVR24): 25-Feb-2011		

AI444014					
Results and Analysis					
Analysis description	**Primary Analysis - Extended Rapid Virologic Response**				
Analysis population and time point description	An analysis of antiviral activity was conducted after all subjects reached Week 12. Response rates and 80% exact binomial CIs were presented by treatment group using modified ITT (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects.				
Descriptive statistics and estimate variability	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	eRVR* Responders (%)	5 (41.7)	10 (83.3)	9 (75.0)	1 (8.3)
	80% CIs	(21.9, 63.8)	(61.4, 95.5)	(52.5, 90.4)	(0.9, 28.7)
Notes	* Undetectable HCV RNA at both Weeks 4 and 12 on-treatment				
Analysis description	**Secondary analysis - Rapid Virologic Response**				
Analysis population and time point description	Secondary binary efficacy endpoints (RVR, EVR. cEVR, EOT, SVR4, SVR12, and SVR24) are assessed with response rates and 80% exact binomial CIs by treatment group using mITT.				
Descriptive statistics and estimate variability	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	RVR* Responders (%)	5 (41.7)	11 (91.7)	10 (83.3)	1 (8.3)
	80% CIs	(21.9, 63.8)	(71.3, 99.1)	(61.4, 95.5)	(0.9, 28.7)
Notes	* Undetectable HCV RNA at Week 4 on-treatment				
Analysis description	**Secondary analysis - Early Virologic Response**				
Descriptive statistics and estimate variability	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	EVR* Responders (%)	9 (75.0)	12 (100)	10 (83.3)	8 (66.7)
	80% CIs	(52.5, 90.4)	(82.5, 100)	(61.4, 95.5)	(44.1, 84.6)
Notes	* Defined as ≥ 2 $\log_{10}$ decrease in HCV RNA from baseline at Week 12 or HCV RNA < 10 IU/mL on-treatment for subjects with baseline HCV RNA < 1000 IU/mL				
Analysis description	**Secondary analysis - Sustained Virologic Response at Week 12**				
Descriptive statistics and estimate variability	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	SVR4* Responders (%)	5 (41.7)	11 (91.7)	10 (83.3)	3 (25.0)
	80% CIs	(21.9, 63.8)	(71.3, 99.1)	(61.4, 95.56)	(9.6, 47.5)
Notes	* Undetectable HCV RNA at follow-up Week 12				

AI444014					
Analysis description	**Secondary analysis - Sustained Virologic Response at Week 24**				
Descriptive statistics and estimate variability	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	SVR24* Responders (%)	5 (41.7)	10 (83.3)	10 (83.3)	3 (25.0)
	80% CIs	(21.9, 63.8)	(61.4, 95.5)	(61.4, 95.5)	(9.6, 47.3)
Notes	* Undetectable HCV RNA at follow-up Week 12				
Analysis description	**Secondary analysis - Virologic Failure**				
Descriptive statistics	Treatment group	Placebo	DCV 3 mg	DCV 10 mg	DCV 60 mg
	Number of subjects	12	12	12	12
	VBT*	0	2	0	1
	Relapse**	5	2	1	1
Notes	*VBT defined as confirmed > 1 $\log_{10}$ increase over nadir or confirmed HCV RNA ≥ LLOQ after confirmed undetectable HCV RNA while on treatment. VBT must be confirmed at the next scheduled assessment. **Relapse: detectable HCV RNA during follow-up after undetectable HCV RNA (< LLOQ, TND) at EOT				
	y) Pre-existing NS5A polymorphisms at amino acid positions associated with resistance were detected by population sequencing in subject samples from all 3 DCV dosing groups. z) Pre-existing NS5A polymorphisms included M28M/V, H58H/P, and E62E/D for HCV GT-1a; and R30Q, L31M/V, Q54H/N/Q/Y, P58A/S/T, Q62E, A92A/E/T/V, and Y93C/H/Y for HCV GT-1b. aa) Of the 11 subjects treated with DCV who met virologic failure, 4 had pre-existing polymorphisms at sites shown to be associated with resistance. bb) Emergent NS5A resistance variants detected in HCV GT-1a subject samples at the time of failure included Q30E/H/R, L31M, H58D, and Y93C. cc) In HCV GT-1b subject samples, emergent NS5A variants detected included L28M, L31M, and Y93H. Emerging NS5A resistance variants were consistent with those variants that have been described previously.				

BMS - Bristol-Myers Squibb, CI(s) - confidence interval(s), DCV - daclatasvir, GT(s) - genotype(s), EOTR - end of treatment response, eRVR - extended rapid virologic response, EVR - early virologic response, HCV - hepatitis C virus, LLOQ - less than the limit of quantitation, mITT - modified intent-to-treat, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR12, 24 - sustained virologic response at follow-up Weeks 12 and 24, respectively, TND - target not detected, VBT - virologic breakthrough

AI444011		
• **Title:** A Phase 2b Study of BMS-790052 in Combination with Peginterferon Alfa-2a and Ribavirin in Chronic Hepatitis C Genotype 1 Infected Subjects Who are Null or Partial Responders to Prior Treatment with Peginterferon Alfa plus Ribavirin Therapy		
Study identifier	AI444011	
Design	Ongoing, randomized, double blinded, Phase 2b study in HCV GT 1-infected patients who failed prior interferon-based therapy (i.e., prior null or prior partial responders): dd) DCV 20 mg or DCV 60 mg QD/pegIFNα/RBV - Prior null responders (1:1) ee) DCV 20 mg or DCV 60 mg or placebo QD/pegIFNα/RBV - Prior partial responders (4:4:1)	
	Duration of main phase	Up to 24 or 48 weeks on-treatment
	Duration of follow-up phase	Up to 24 or 48 weeks follow-up
Hypothesis	**Primary:** In chronically infected HCV GT-1 subjects who failed prior interferon-based therapy, at least 1 dose of DCV combined with pegIFNα-2a/RBV can be identified which is safe, well-tolerated, and demonstrates eRVR rates: ff) > 25% among prior null responders, and gg) more than 35% > control (placebo/pegIFNα-2a/RBV) among prior partial responders **Co-primary:** In chronically infected HCV GT-1 subjects who failed prior interferon-based therapy, at least 1 dose of DCV combined with pegIFNα-2a/RBV can be identified which is safe, well-tolerated, and demonstrates SVR24 rates: hh) > 20% among prior null responders, and ii) more than 20% > control (placebo/pegIFNα-2a/RBV) among prior partial responders	
Treatment groups	419 subjects were randomized and treated: 203 [133 prior null responders and 70 prior partial responders], 199 [132 prior null responders and 67 prior partial responders], and 17 subjects in the DCV 20 mg/pegIFN [placebo/pegIFN □/RBV groups, respectively.	
	Prior Null Responders DCV 20 mg	Prior null responders received DCV 20 mg QD/pegIFNα/RBV up to 24 weeks. Before the Week 24 visit, subjects who achieved the PDR, underwent a second randomization (1:1) to either: jj) Stop all therapy, or kk) An additional 24 weeks of treatment with pegIFNα/RBV Subjects who did not achieve the PDR were administered an additional 24 weeks of pegIFNα/RBV, for a total of 48 weeks of therapy.
	Prior Null Responders DCV 60 mg	Prior null responders received DCV 60 mg QD/pegIFNα/RBV up to 24 weeks. Before the Week 24 visit, subjects who achieved the PDR, underwent a second randomization (1:1) to either: ll) Stop all therapy, or mm) An additional 24 weeks of treatment with pegIFNα/RBV Subjects who did not achieve the PDR received an additional 24 weeks of pegIFNα/RBV, for a total of 48 weeks of therapy.

AI444011			
	Prior Partial Responders DCV 20 mg		Prior partial responders received DCV 20 mg QD/pegIFNα/RBV up to 24 weeks. Before the Week 24 visit, subjects who achieved the PDR, underwent a second randomization (1:1) to either: nn) Stop all therapy, or oo) An additional 24 weeks of treatment with pegIFNα/RBV Subjects who did not achieve the PDR received an additional 24 weeks of pegIFNα/RBV, for a total of 48 weeks of therapy.
	Prior Partial Responders DCV 60 mg		Prior partial responders received DCV 60 mg QD/pegIFNα/RBV up to 24 weeks. Before the Week 24 visit, subjects who achieved the PDR, underwent a second randomization (1:1) to either: pp) Stop all therapy, or qq) An additional 24 weeks of treatment with pegIFNα/RBV Subjects who did not achieve the PDR received an additional 24 weeks of pegIFNα/RBV, for a total of 48 weeks of therapy.
	Prior Partial Responders Placebo		Prior partial responders received placebo QD/pegIFNα/RBV up to 24 weeks. All subjects randomized to placebo, regardless of PDR status, received pegIFNα/RBV for an additional 24 weeks, for a total of 48 weeks of therapy.
Endpoints and definitions	Primary endpoints	Co-primary endpoint	Proportion of subjects in each cohort (prior partial responders and prior null responders) with eRVR, defined as undetectable HCV RNA at both Weeks 4 and 12
		Co-Primary endpoint	Proportion of subjects in each cohort (prior partial responders and prior null responders) with SVR24, defined as undetectable HCV RNA at follow-up Week 24
	Secondary endpoints	Secondary endpoint	Proportion of subjects in each cohort (prior partial responders and prior null responders) with RVR, defined as undetectable HCV RNA at Week 4
		Secondary endpoint	Proportion of subjects in each cohort (prior partial responders and prior null responders) with cEVR, defined as undetectable HCV RNA at Week 12
		Secondary endpoint	Proportion of subjects in each cohort (prior partial responders and prior null responders) with SVR12, defined as undetectable HCV RNA at follow-up Week 12
Database lock	07-Mar-2013		

AI444011						
Results and Analysis						
Analysis description	**Co-Primary Analysis - Extended Rapid Virologic Response**					
Analysis population and time point description	Response rates and 80% CIs were presented by treatment regimen using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects. For eRVR, the difference in the proportions of prior partial responders with eRVR between each DCV regimen and the placebo regimen was presented using mITT with a difference estimate (DCV - PBO) and CI. The CI was based on a normal approximation to the binomial distribution using unpooled proportions to compute the standard error of the difference.					
Descriptive statistics and estimate variability	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	eRVR* (Responder; %)	24/133(18.0)	26/132 (19.7)	18/70 (25.7)	24/67 (35.8)	0/17
	80% CIs	(13.8, 22.3)	(15.3, 24.1)	(19.0, 32.4)	(28.3, 43.3)	(0.0, 0.0)
	Difference: DCV - Placebo (80% CIs)	---	---	25.7 (19.0, 32.4)	35.8 (28.3, 43.3)	---
Notes	* Undetectable HCV RNA (< LLOQ, TND) at both Weeks 4 and 12 on-treatment					
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					
Analysis description	**Co-Primary Analysis - Sustained Virologic Response at Follow-up Week 24**					
Descriptive statistics and estimate variability	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	SVR24* (Responder; %)	25/133 (18.8)	29/132 (22.0)	17/70 (24.3)	29/67 (43.3)	0/17
	80% CIs	(14.5, 23.1)	(17.4, 26.6)	(17.7, 30.9)	(35.5, 51.0)	(0.0, 0.0)
Notes	* Undetectable HCV RNA (< LLOQ, TND) at Follow-up Week 24					
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					

AI444011						
Analysis description	**Secondary Analysis - Rapid Virologic Response**					
Analysis population and time point description	Response rates and 80% CIs were presented by treatment regimen using mITT The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects.					
Descriptive statistics and estimate variability	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	RVR* (Responder; %)	29/133(21.8)	28/132 (21.2)	18/70 (25.7)	26/67 (38.8)	0/17
	80% CIs	(17.2, 26.4)	(16.0, 25.0)	(19.0, 32.4)	(31.2, 46.4)	(0.0, 0.0)
Notes	* Undetectable HCV RNA at Week 4					
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					
Analysis description	**Secondary Analysis - Complete Early Virologic Response**					
Descriptive statistics and estimate variability	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	cEVR* (Responder; %)	40/133(30.1)	45/132 (34.1)	31/70 (44.3)	38/67 (56.7)	0/17
	80% CIs	(23.0, 35.2)	(28.8, 39.4)	(36.7, 51.9)	(49.0, 64.5)	(0.0, 0.0)
Notes	* Undetectable HCV RNA at Week 12					
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					
Analysis description	**Secondary Analysis - Sustained Virologic Response at Follow-up Week 12**					
Descriptive statistics and estimate variability	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	SVR12* (Responder; %)	26/133 (19.5)	31/132 (23.5)	18/70 (25.7)	32/67 (47.8)	0/17
	80% CIs	(15.1, 24.0)	(18.8, 28.2)	(19.0, 32.4)	(39.9, 55.6)	(0.0, 0.0)
Notes	* Undetectable HCV RNA at follow-up Week 12					
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					

AI444011						
Analysis description	**Secondary Analysis - Virologic Failure**					
Descriptive statistics	Treatment group	Prior Null Responders		Prior Partial Responders		
		DCV 20 mg	DCV 60 mg	DCV 20 mg	DCV 60 mg	Placebo
	Number of subjects	133**	132***	70	67	17
	VBT[#]	36.1 (48)	41.7 (55)	32.9 (23)	26.9 (18)	5.9 (1)
	Relapse Rate[##]	44.4 (20/45)	37.5 (18/48)	33.3 (9/27)	30.0 (12/40)	75.0 (3/4)
	** N = 134 randomized subjects. One randomized subject never received study drug.					
	*** N = 133 randomized subjects. One randomized subject never received study drug.					
	[#]Confirmed > 1 $\log_{10}$ increase in HCV RNA over nadir or confirmed HCV RNA after confirmed undetectable HCV RNA. Measurements should be confirmed at the next scheduled visit.					
	[##]Detectable HCV RNA during follow-up after undetectable HCV RNA at EOT					

NS5A resistance associated polymorphisms (RAPs) were detected in 32% (118/374) of subjects:

- GT-1a (N = 247):
  - 36 of 247 subjects had baseline NS5A RAPs; GT-1a samples included methionine (M)28 leucine (L)/threonine (T)/valine (V), glutamine (Q)30 histidine (H), L31M, H54 tyrosine (Y), H58 cysteine (C)/aspartate (D)/asparagine (N)/proline (P)/Q, glutamate (E)62D, and Y93C
- GT-1b (N = 127):
  - 82 of 127 subjects had baseline NS5A RAPs; GT-1b samples included L28M/V, arginine (R)30H/Q, L31M, Q54H/N/Y, P58A/Q/Serine (S), Q62E/lysine (K)/N/R/S, alanine (A)/92T/V, and Y93 phenylalanine (F)/H

The most prevalent baseline NS5A RAP in subjects with GT-1a was L31M, detected in 25% (9/36) of subjects; 6 of 9 were prior null responders and 3 of 9 were prior partial responders.100% (9/9) of subjects with the L31M RAP failed treatment. The most prevalent baseline NS5A RAP in subjects with GT-1b was Q54H, detected in 59% (48/82) of subjects; 33 of 48 were prior null responders and 15 of 48 were prior partial responders. 65% (31/48) of subjects with the Q54H RAP failed treatment.

Analysis of the effects of pre-existing signature DCV-resistant variants indicated there may be an association between GT-1a NS5A RAPs (M28V/L/T, L31M, H58C/D/N/P/Q, and Y93C) and virologic failure since 96% (25/26) of subjects with these variants failed treatment.

Of GT-1a virologic failures, emergent substitutions at M28A/glycine (G)/S/T/V, Q30D/E/G/H/K/N/R/T, L31 isoleucine (I)/M/V, H54R/Y, H58D/N/P/Q/V, A92P, and Y93C/H/N/R/S were detected. Q30 variants were detected most frequently either alone or in combination with other NS5A RAVs at amino acid positions 28, 31, 58, and 93 (91%; 180/197 failures). Of GT-1b virologic failures, emergent substitutions at L28M, P29X, R30H/K/L/P/Q/R/S, L31F/I/M/V, P32X, Q54H/Y, P58S, A92E/K/T, and Y93H were detected. Y93H combined with variants at L31 (L31I/M/V) predominated and was detected in 81% (57/70) of GT-1b failures with NS5A sequence.

Replacement or partial replacement of emergent NS5A RAPs was observed in subjects when monitored out to follow-up Week 48. Of the 148 subjects with GT-1a examined at follow-up Week 48, replacement or partial replacement of these NS5A variants was observed in 2% (3/148) and 25% (36/148) of subjects, respectively; in 2 subjects who

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	relapsed with emergent Y93H, reversion/outgrowth by baseline sequence was observed.

BMS - Bristol-Myers Squibb, CI(s) - confidence interval(s), DCV - daclatasvir, GT(s) - genotype(s), EOTR - end of treatment response, eRVR - extended rapid virologic response, EVR - early virologic response, HCV - hepatitis C virus, LLOQ - less than the limit of quantitation, mITT - modified intent-to-treat, pegIFNα - peginterferon alfa, QD - once daily, RAPs - resistance associated polymorphisms, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR12, 24 - sustained virologic response at follow-up Weeks 12 and 24, respectively, TND - target not detected, VBT - virologic breakthrough

AI444031			
• **Title:** A Phase 2b Pilot Study of Short-Term Treatment of BMS-790052 in Combination with Peg-Interferon Alfa-2a and Ribavirin in Treatment Naive Subjects with Chronic Hepatitis C Genotype 2 or 3 Infection			
Study identifier	AI444031		
Design	Phase 2b, randomized, placebo-controlled, response-guided study in treatment naive subjects with HCV GT-2 or GT-3. Subjects were randomized 1:1:1 to receive either rr) DCV 60 mg QD/pegIFNα/RBV for 12 weeks ss) DCV 60 mg QD/pegIFNα/RBV for 16 weeks tt) Placebo/pegIFNα/RBV for 24 weeks		
	Duration of main phase	12, 16, or 24 weeks	
	Duration of follow-up phase	24, 32, or 48 weeks	
Hypothesis	For treatment-naive subjects chronically-infected with HCV GT-2 or -3, a shorter duration of antiviral therapy (12 or 16 weeks) of DCV combined with pegIFN □2a/RBV can be identified which is safe and well tolerated, and has observed efficacy comparable to 24 weeks of pegIFN □2a/RBV.		
Treatment groups	151 subjects were treated: 50 with DCV 60 mg/pegIFN [60 mg/pegIFN □/RBV f		
	DCV 60 mg/pegIFN □1/2RB week group	• Subjects who achieved a PDR (defined as HCV RNA < LLOQ, target detected [TD] or TND at Week 4 and HCV RNA at < LLOQ, TND at Week 10) completed 12 weeks of DCV/pegIFN [randomization and proceeded to post-treatment follow-up. • Subjects who did not achieve a PDR were required to receive 24 weeks of therapy. At Week 12 of DCV/pegIFN [treatment, these subjects received an additional 12 weeks of placebo/pegIFN □/RBV.	
	DCV 60 mg/pegIFN □1/6RB week group	• Subjects who achieved a PDR (defined as HCV RNA < LLOQ, target detected [TD] or TND at Week 4 and HCV RNA at < LLOQ, TND at Week 10) completed 16 weeks of DCV/pegIFN□/RBV therapy based on their initial randomization and proceeded to post-treatment follow-up. uu) Subjects who did not achieve a PDR were required to receive 24 weeks of therapy. At Week 12 of DCV/pegIFN [treatment, these subjects received an additional 12 weeks of placebo/pegIFN □/RBV.	
	Placebo/pegIFN □ V	vv) Subjects in this group received 24 weeks of placebo/pegIFN [therapy	
Endpoints and definitions	Primary endpoint	Primary endpoint	Proportion of subjects for each HCV GT with SVR24, defined as HCV RNA < LLOQ, TND at follow-up Week 24.
	Secondary endpoints	Secondary endpoint	Proportion of subjects for each HCV GT with RVR: HCV RNA < LLOQ, TND at Week 4
		Secondary endpoint	Proportion of subjects for each HCV GT with SVR12: HCV RNA < LLOQ, TND at follow-up Week 12
		Secondary endpoint	Frequency of genotypic substitutions associated with virologic failure for each HCV GT
Database lock	19-Oct-2012		

AI444031							
Results and Analysis							
Analysis description	**Primary Analysis - SVR24**						
Analysis population and time point description	Response rates and 80% CIs were presented by treatment regimen and GT using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects. For the primary endpoint (SVR24), the difference in the proportions of subjects with antiviral response between each DCV treatment regimen and the placebo regimen was presented for each HCV GT using modified ITT with a difference estimate (DCV - placebo) and 80% CI. The CI was based on a normal approximation to the binomial distribution using unpooled proportions to compute the standard error of the difference.						
Descriptive statistics and estimate variability	Treatment group	GT-2			GT-3		
		DCV 12-week	DCV 16-week	Placebo 24-week	DCV 12-week	DCV 16-week	Placebo 24-week
	Number of subjects	24	23	24	26	27	27
	SVR24* (Responder; %)	20 (83.3)	19 (82.6)	15 (62.5)	18 (69.2)	18 (66.7)	16 (59.3)
	80% CIs	73.6, 93.1	72.5, 92.7	49.8, 75.2	57.6, 80.8	55.0, 78.3	47.1, 71.4
	Difference: DCV - Placebo (80% CIs)	20.8 (4.9, 36.8)	20.1 (3.9, 36.3)	-	10.0 (-6.8, 26.7)	7.4 (-9.4, 24.2)	-
Notes	* HCV RNA < LLOQ, TND at follow up Week 24						

AI444031							
Analysis description	**Secondary Analysis - RVR**						
Analysis population and time point description	Response rates and 80% CIs were presented by treatment regimen and GT using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects.						
Descriptive statistics and estimate variability	Treatment group	GT-2			GT-3		
		DCV 12-week	DCV 16-week	Placebo 24-week	DCV 12-week	DCV 16-week	Placebo 24-week
	Number of subjects	24	23	24	26	27	27
	RVR* (Responder; %)	21 (87.5)	17 (73.9)	10 (41.7)	22 (84.6)	20 (74.1)	10 (37.0)
	80% CIs	78.8, 96.2	62.2, 85.6	28.8, 54.6	75.5, 93.7	63.3, 84.9	25.1, 48.9
Notes	* HCV RNA < LLOQ, TND at Week 4						
Analysis description	**Secondary Analysis - cEVR**						
Analysis population and time point description	Response rates and 80% CIs were presented by treatment regimen and GT using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects.						
Descriptive statistics and estimate variability	Treatment group	GT-2			GT-3		
		DCV 12-week	DCV 16-week	Placebo 24-week	DCV 12-week	DCV 16-week	Placebo 24-week
	Number of subjects	24	23	24	26	27	27
	cEVR* (Responder; %)	22 (91.7)	19 (82.6)	18 (75.0)	21 (80.8)	24 (88.9)	16 (59.3)
	80% CIs	84.4, 98.9	72.5, 92.7	63.7, 86.3	70.9, 90.7	81.1, 96.6	47.1, 71.4
Notes	* HCV RNA < LLOQ, TND at Week 12						

AI444031							
Analysis description	**Secondary Analysis -** Frequency of genotypic substitutions associated with virologic failure for each HCV GT						
Descriptive statistics	Treatment group	GT-2			GT-3		
		DCV 12-week	DCV 16-week	Placebo 24-week	DCV 12-week	DCV 16-week	Placebo 24-week
	Number of subjects	24	23	24	26	27	27
	VBT	0	1 (4.3)	1 (4.2)	0	0	1 (3.7)
	Relapse	1/23 (4.3)	0/21	2/22 (14.3)	6/25 (24.0)	6/24 (25.0)	3/21 (14.3)
Notes	* VBT: Confirmed > 1 $\log_{10}$ increase in HCV RNA over nadir or confirmed HCV RNA after confirmed HCV RNA < LLOQ, TND, while on treatment. Measurements should be confirmed within 2 weeks of receipt of initial HCV RNA measurement or at the next scheduled assessment, whichever was sooner **Relapse: HCV RNA ≥ LLOQ after confirmed HCV RNA < LLOQ, TND during TND at EOT.						

AI444031	
	GT-2 Sequence analysis of baseline samples from 44 of 47 GT-2 subjects revealed that NS5A polymorphisms previously shown to confer resistance to DCV in GT-1 NS5A sequences were detected in 52% (23/44) of these subjects (NS5A-L31M). Only 4 of the 23 GT-2 subjects with NS5A-L31M did not achieve SVR24. 2 of the 47 GT-2 subjects (1 with GT-2a and 1 with GT-2b) had an HCV RNA level □ 1000 IU/mL at treatment Week 1. ww) One subject (GT-2a, IL-28B rs12979680 CC genotype) experienced a slow viral load decline during the first 12 weeks of treatment; this subject achieved SVR24. No emergent DCV-resistant variants were detected in the first 8 weeks of treatment although a pre-existing DCV-resistant variant (NS5A-L31M) was detected throughout this period. NS5A-L31M was also detected in 22 other GT-2 subjects on the study. xx) The other subject (GT-2b) experienced rapid VBT. Resistance analysis revealed the emergence of NS5A-Y93H at Week 2 of treatment followed by an additional substitution at NS5A-N62 by Week 4. By Week 12, HCV RNA for this subject was < LLOQ, TND. This subject who had no pre-existing NS5A polymorphisms associated with DCV resistance and carried the IL-28B CC genotype, received 16 weeks of treatment, and subsequently achieved SVR24. **GT-3** Sequence analysis of baseline samples from 52 of 53 GT-3 subjects revealed that NS5A polymorphisms previously shown to confer resistance to DCV in GT-1 NS5A sequences were detected in 15% (8/52) of these subjects (NS5A-A30K/ valine (V) and/or NS5A-Y93H). Half (4/8) experienced relapse and half (4/8) ultimately achieved SVR24. 9 GT-3 subjects had an HCV RNA level □1000 IU/mL at treatment Week 1: yy) 3/9 subjects achieved SVR24: 1 had a pre-existing NS5A polymorphism associated with DCV resistance (NS5A-Y93H) and carried the IL-28B CT genotype and 2 had no detectable baseline NS5A polymorphisms and carried the IL-28B CC or CT genotypes. zz) 6/9 subjects failed treatment: 3 had pre-existing NS5A polymorphisms (NS5A-Y93H or NS5A-A30K) that confer resistance to DCV and 3 (all with IL28B CC genotype) had no detectable pre-existing DCV polymorphisms, but an emergence of the NS5A-Y93H substitution. Of the 4 GT-3 subjects with virologic failure during treatment, 3/4 had an emergence of NS5A-Y93H. All 12 GT-3 relapsers had NS5A-A30K or NS5A-Y93H resistance variants detected at virologic failure: aaa) NS5A-A30K was detected in 2 subjects bbb) NS5A-Y93H was detected in 10 subjects

BMS - Bristol-Myers Squibb, cEVR - complete early virologic response, CI(s) - confidence interval(s), DCV - daclatasvir, GT - genotype, HCV - hepatitis C virus, ITT - intent-to-treat, mITT - modified intent-to-treat, PDR - protocol defined response, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR12 - sustained virologic response at follow-up Week 12, SVR24 - sustained virologic response at follow-up Week 24, TND - target not detected, VBT - virologic breakthrough

AI444042	
• **Title:** A Phase 3 Evaluation of Daclatasvir (BMS-790052) in Combination with Peg-Interferon Alfa-2a and Ribavirin in Treatment-Naive Subjects with Chronic Hepatitis C Genotype 4	
Study identifier	AI444042

AI444042			
Design	Phase 3, randomized, double-blind study in treatment naive subjects with HCV GT-4. Subjects were randomized 2:1 to receive either ccc) DCV 60 mg QD/pegIFNα/RBV for 24 or 48 weeks based on response ddd) Placebo/pegIFNα/RBV for 48 weeks		
	Duration of main phase	24 or 48 weeks	
	Duration of follow-up phase	24 or 48 weeks	
Hypothesis	In treatment-naive subjects chronically-infected with HCV GT-4, BMS-790052 (daclatasvir) in combination with pegIFN □2a and RBV is safe and demonstrates SVR12 (defined as HCV RNA < LOQ [25 IU/mL] at post-treatment Week 12) rates greater than pegIFN □2a/RBV alone.		
Treatment groups	124 subjects were treated: 82 with DCV 60 mg/pegIFN □/RBV and placebo/pegIFN □/RBV		
	DCV 60 mg/pegIFN □/RBV	• Subjects who achieved a VR(4&12) (defined as HCV RNA undetectable [< LLOQ, TND] at both Weeks 4 and 12) completed therapy at Week 24 and were followed for an additional 48 weeks of post-treatment follow-up. • Subjects who did not achieve a VR(4&12) received 48 weeks of therapy, and were followed for 24 weeks of post-treatment follow-up.	
	Placebo/pegIFN □/RBV	eee) Subjects received 48 weeks of therapy, and were followed for 24 weeks of post-treatment follow-up.	
Endpoints and definitions	Primary endpoint	Primary endpoint	Proportion of subjects with SVR12, defined as HCV RNA < LLOQ, TD or TND at follow-up Week 12.
	Secondary endpoint	Secondary endpoints	• Proportion of subjects who achieved HCV RNA < LLOQ at Weeks 1, 2, 4, 6, 8, and 12; at both Weeks 4 and 12; at end of treatment (EOT, up to 48 weeks); post-treatment Week 24 (SVR24); or post-treatment Week 48 for subjects who achieved virologic response (HCV RNA undetectable [< LLOQ, TND]) at both Weeks 4 and 12 (VR[4&12]) • Proportion of subjects who achieved HCV RNA undetectable at Weeks 1, 2, 4, 6, 8, and 12; at both Weeks 4 and 12; at EOT (up to 48 weeks); post-treatment Week 12; post-treatment Week 24; or post-treatment Week 48 for subjects who achieved VR(4&12)
Database lock	18-Dec-2013		
Results and Analysis			
Analysis description	**Primary Analysis - SVR12**		
Analysis population and time point description	Response rates and 95% CIs were presented by treatment regimen using modified intent-to-treat (mITT). The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects. For the primary endpoint (SVR12), the difference in the proportions of subjects with antiviral response between the DCV treatment regimen and the placebo regimen was presented using mITT with a difference estimate (DCV - placebo) and 95% CI. The CI was based on a normal approximation to the binomial distribution using unpooled proportions to compute the standard error of the difference.		
Descriptive statistics and estimate	Treatment group	DCV/pegIFNα/RBV	Placebo/pegIFNα/RBV
	Number of subjects	82	42

AI444042			
variability	SVR12* (Responder; %)	60 (73.2)	16 (38.1)
	95% CIs	63.6, 82.8	23.4, 52.8
	Difference: DCV - Placebo (95% CIs)	35.1 (17.5, 52.6)	-
	SVR12 with imputation**	67 (81.7)	18 (42.9)
	95% CIs	73.3, 90.1	27.9, 57.8
	Difference: DCV - Placebo (95% CIs)	38.9 (21.7, 56.0)	-
Notes	* HCV RNA < LLOQ, TD or TND at follow-up Week 12 **Subjects with missing HCV RNA at follow-up Week 12 were counted as SVR12 responders if they had HCV RNA < LLOQ, TD or TND at the next available measurement		

AI444042					
Analysis description	**Secondary Analyses -** • HCV RNA < LLOQ at Weeks 1, 2, 4, 6, 8, and 12; at both Weeks 4 and 12; at end of treatment (EOT, up to 48 weeks); post-treatment Week 24 (SVR24); or post-treatment Week 48 for subjects who achieved virologic response (HCV RNA undetectable [< LLOQ, TND]) at both Weeks 4 and 12 (VR[4&12]) • HCV RNA undetectable at Weeks 1, 2, 4, 6, 8, and 12; at both Weeks 4 and 12; at EOT (up to 48 weeks); post-treatment Week 12; post-treatment Week 24; or post-treatment Week 48 for subjects who achieved VR(4&12)				
Analysis population and time point description	Response rates and 95% CIs were presented by treatment regimen using mITT. The numerator was based on subjects meeting the response criteria. The denominator was based on all treated subjects.				
Descriptive statistics	Treatment group	DCV/pegIFNα/RBV		Placebo/pegIFNα/RBV	
	Endpoint	HCV RNA < LLOQ, TD or TND	HCV RNA < LLOQ, TND	HCV RNA < LLOQ, TD or TND	HCV RNA < LLOQ, TND
	Week 1	44 (53.7)	12 (14.6)	2 (4.8)	0
	Week 2	73 (89.0)	37 (45.1)	5 (11.9)	4 (9.5)
	Week 4	75 (91.5)	70 (85.4)	8 (19.0)	5 (11.9)
	Week 6	69 (84.1)	66 (80.5)	17 (40.5)	7 (16.7)
	Week 8	72 (87.8)	72 (87.8)	20 (47.6)	16 (38.1)
	Week 12	70 (85.4)	69 (84.1)	25 (59.5)	20 (47.6)
	Weeks 4 and 12	69 (84.1)	65 (79.3)	8 (19.0)	5 (11.9)
	EOT	78 (92.7)	74 (90.2)	27 (64.3)	27 (64.3)
	Follow-up Week 12	60 (73.2)	56 (68.3)	16 (38.1)	16 (38.1)

CI(s) - confidence interval(s), DCV - daclatasvir, EOT - end of treatment, GT - genotype, HCV - hepatitis C virus, mITT - modified intent-to-treat, LLOQ - lower limit of quantitation, LOQ - limit of quantitation, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, SVR12 - sustained virologic response at follow-up Week 12, SVR24 - sustained virologic response at follow-up Week 24, TD - target detected, TND - target not detected, VR - virologic response

AI447026			
• **Title:** A Phase 3 Japanese Study of BMS-790052 plus BMS-650032 Combination Therapy in Chronic Hepatitis C Genotype 1b Infected Subjects Who are Non Response to Interferon plus Ribavirin and Interferon Based Therapy Ineligible naïve /intolerant			
Study identifier	AI447026		
Design	Open-label, Phase 3 study, in 2 parallel Japanese populations: non-responder (null and partial responder) and IFN-based therapy ineligible /intolerant subjects infected with HCV GT-1b		
	Duration of main phase		Open-label, DCV/ASV Dual therapy up to 24 weeks for both populations
	Duration of Follow-up phase		24 weeks follow-up for both populations
	Rescue therapy		Non-responders who met the criteria were considered treatment failure of DAAs and could be administered a rescue therapy of DCV/ASV/peg IFN/RBV Quad therapy for up to 24 additional weeks and followed post-treatment for 24 weeks, regardless of HCV RNA status at EOT
Hypothesis	Co-administration of DCV/ASV for 24 weeks for HCV GT-1b infection can achieve SVR24 rate whose lower bound of the estimated 95% CI is > 45% for non-responder and > 30% for IFN-based therapy ineligible naïve/intolerant subjects		
Treatment groups	Non-responder		DCV 60 mg QD/ASV 100 mg BID Dual therapy for up to 24 weeks and followed post-treatment for 24 weeks, regardless of HCV RNA status at EOT
	IFN-based therapy ineligible naïve/intolerant		DCV 60 mg QD/ASV 100 mg BID Dual therapy for up to 24 weeks and followed post-treatment for 24 weeks, regardless of HCV RNA status at EOT.
Endpoints and definitions	Primary endpoint	Primary endpoint	Proportion of subjects with SVR24, defined as HCV RNA below LLOQ (< 15 IU/mL), target detected (TD) or target not detected (TND) at Week 24 post-treatment for each population separately
	Secondary endpoint	Secondary endpoint(s)	fff) Proportion of subjects who achieved HCV RNA below LLOQ, TD or TND at Weeks: 1, 2, 4, 6, 8, 10, and 12; Weeks: 4 and 12; EOT, or post-treatment follow-up Week 12 ggg) Proportion of subjects who achieved HCV RNA below LLOQ, TND at Weeks: 1, 2, 4, 6, 8, 10, and 12; Weeks: 4 and 12; EOT, or post-treatment Week 12, post-treatment Week 24 hhh) Proportion of subjects with SVR24 by IL28B status (CC, CT, or TT genotype at the IL28B rs12979860)
Database lock	10-May-2013		

AI447026			
Results and Analysis			
Analysis description	**Primary Analysis - Sustained Virologic Response at Follow-up Week 24**		
Analysis population and time point description	Primary efficacy population was based on the modified intent-to-treat (mITT) population where the numerator was based on subjects who met the response criteria (at post-treatment Week 24). The denominator was based on all treated subjects.		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	SVR24* (Responder; %)	70/87 (80.5)	118/135 (87.4)
	95% CIs	(72.1, 88.8)	(81.8, 93.0)
Notes	*Defined as HCV RNA < LLOQ (< 15 IU/mL) TD or TND at post treatment Week 24		
Analysis description	**Secondary analysis - HCV RNA Below LLOQ, TD or TND at Week 4**		
Analysis population and time point description	Secondary analyses were based on the modified intent-to-treat (mITT) population where the numerator was based on subjects who met the response criteria. The denominator was based on all treated subjects at visit weeks defining the endpoint.		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	Week 4 (Responder; %)	80/87 (92.0)	132/135 (97.8)
	95% CIs	(86.2, 97.7)	(95.3, 100.0)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL) TD or TND at Week 4		
Analysis description	**Secondary analysis - HCV RNA Below LLOQ, TD or TND at Week 12**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	Week 12 (Responder; %)	78/87 (89.7)	125/135 (92.6)
	95% CIs	(83.3, 96.1)	(88.2, 97.0)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL) TD or TND at Week 12		
Analysis description	**Secondary analysis - HCV RNA Below LLOQ, TD or TND at Week 24**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	Week 24 (Responder; %)	75/87 (86.2)	120/135 (88.9)
	95% CIs	(79.0, 93.5)	(83.6, 94.2)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL) TD or TND at Week 24		

AI447026			
Analysis description	**Secondary analysis - HCV RNA Below LLOQ TD or TND at follow-up Week 12**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	Week 24 (Responder; %)	70/87 (80.5)	119/135 (88.1)
	95% CIs	(72.1, 88.8)	(82.7, 93.6)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL), TD or TND at follow-up Week 12		
Analysis description	**Secondary analysis - RVR**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	RVR (Responder; %)	53/87 (60.9)	114/135 (84.4)
	95% CIs	(50.7, 71.2)	(78.3, 90.6)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL), TND at Week 4		
Analysis description	**Secondary analysis - cEVR**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	cEVR (Responder; %)	77/87 (88.5)	125/135 (92.6)
	95% CIs	(81.8, 95.2)	(88.2, 97.0)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL), TND at Week 12		
Analysis description	**Secondary analysis - eRVR**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	eRVR (Responder; %)	48/87 (55.2)	106/135 (78.5)
	95% CIs	(44.7, 65.6)	(71.6, 85.4)
Analysis description	**Secondary analysis - SVR12 (HCV RNA < LLOQ, TND)**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	Number of subjects	87	135
	SVR12 (Responder; %)	70/87 (80.5)	119/135 (88.1)
	95% CIs	(72.1, 88.8)	(82.7, 93.6)
Notes	*Defined as HCV RNA below LLOQ (< 15 IU/mL), TND at follow-up Week 12		

AI447026			
Analysis description	**Secondary analysis - Sustained Virologic Response at Post-treatment Week 24 by IL-28B rs12979860 Status**		
Descriptive statistics and estimate variability	Treatment group	Non-responder	Ineligible naïve/intolerant
	IL-28B rs12979860 CC	14/16 (87.5)	79/94 (84.0)
	IL-28B rs12979860 CT	52/66 (78.8)	38/40 (95.0)
	IL-28B rs12979860 TT	4/5 (80.0)	1/1 (100.0)

ASV - asunaprevir, BID - twice daily, BMS - Bristol-Myers Squibb, cEVR - complete early virologic response, CI(s) - confidence interval(s), DAA(s) - direct antiviral agent(s), DCV - daclatasvir, EOT - end of treatment, eRVR - extended rapid virologic response, GT(s) - genotype(s), HCV - hepatitis C virus, IFN - interferon, LLOQ - lower limit of quantification, mITT - modified intent-to-treat, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR24 - sustained virologic response at follow-up Week 24, TD - target detected, TND - target not detected

AI447017			
• **Title:** A Phase 2a Study of BMS-790052 and BMS-650032 in Combination Therapy with Japanese Subjects with Genotype 1 Chronic Hepatitis C (HCV) Virus			
Study identifier	AI447017		
Design	Open-label, Phase 2a study in Japanese subjects who were prior null responders to pegIFNα/RBV therapy (Cohorts 1 & 2) or IFN (IFN: includes both the pegylated and non-pegylated forms)/RBV ineligible - naïve/intolerant subjects (Cohorts 3 & 4). The study was conducted in 2 parts: iii) Part 1: Study initiated with a sentinel cohort of 10 prior null responders (Part 1 Cohort 1) to evaluate the safety of the DCV/ASV Dual therapy jjj) Part 2: Review of the Week 4 safety data of all subjects in Cohort 1 allowed the expansion of the study to include Part 2 Cohort 2 (additional prior null responders) and Cohorts 3 & 4 (IFN/RBV ineligible- naïve/intolerant subjects)		
	Duration of main phase	Open-label, DCV/ASV treatment period up to 24 weeks for all subjects	
	Duration of Follow-up phase	48 or 72 weeks follow-up for all cohorts; virologic failures were to be followed through post-treatment Week 12	
	Rescue therapy phase	Prior null responders in Cohorts 1 & 2, who failed treatment, received a rescue therapy of DCV/ASV/pegIFNα/RBV Quad therapy for up to an additional 48 weeks and these subjects were followed for 24 or 48 weeks	
Hypothesis	The observed proportion of null-responder or SOC ineligible naïve/intolerant subjects achieving sustained virologic response at 12 weeks post-treatment (SVR12) (i.e., HCV RNA below the LLOQ at follow-up Week 12) is ≥ 20%.		
Treatment groups	Null responder - sentinel (Cohort 1)	kkk) Sentinel subjects received DCV/ASV for up to 24 weeks lll) Subjects were initially administered DCV 60 mg QD/ASV 600 mg BID; however, based on elevated transaminases noted in an ASV dose-finding study (AI447016), subjects in Cohort 1 had their ASV dose reduced to 200 mg BID after 12 to 20 weeks of treatment mmm) Prior null responders, who failed treatment, were to be administered a rescue therapy of DCV/ASV/pegIFNα/RBV for up to an additional 48 weeks	
	Null responder - expansion (Cohort 2)	nnn) External review of Week 4 safety data of all subjects in Cohort 1 allowed the expansion of the study to Part 2 Cohort 2. ooo) Subjects were administered DCV 60 mg QD/ASV 200 mg BID for up to 24 weeks ppp) Prior null responders, who failed treatment, were to be administered a rescue therapy of DCV/ASV/pegIFNα/RBV for up to an additional 48 weeks	
	IFN/RBV ineligible - naïve/intolerant expansion (Cohorts 3 & 4)	qqq) External review of Week 4 safety data of all subjects in Cohort 1 allowed the expansion of the study to Part 2 Cohorts 3 & 4. rrr) Subjects were administered DCV 60 mg QD/ASV 200 mg BID for up to 24 weeks	
Endpoints and definitions	Primary endpoint	Primary endpoint	Proportion of subjects with SVR12, defined as HCV RNA below LLOQ target detected (TD) or target not detected (TND) at follow-up Week 12

<table>
<tr><td colspan="5">AI447017</td></tr>
<tr><td></td><td>Secondary endpoint</td><td>Secondary endpoint(s)</td><td colspan="2">sss) Proportion of subjects with RVR, defined as HCV RNA < LLOQ (TND) at Week 4
ttt) Proportion of subjects with eRVR, defined as HCV RNA < LLOQ (TND) at both Weeks 4 and 12
uuu) Proportion of subjects with SVR24, defined as HCV RNA < LLOQ (TD or TND) at follow-up Week 24
vvv) Frequency of viral genotypic substitutions associated with virologic failure</td></tr>
<tr><td>Database lock</td><td colspan="4">18-Jun-2012</td></tr>
<tr><td colspan="5"><u>Results and Analysis</u></td></tr>
<tr><td>Analysis description</td><td colspan="4">Primary Analysis - Sustained Virologic Response at Follow-up Week 12 (HCV RNA < LLOQ, TD or TND at follow-up Week 12)</td></tr>
<tr><td>Analysis population and time point description</td><td colspan="4">These analyses were based on all treated subjects (i.e., those in the sentinel or expanded cohort). In general, response rates for binary endpoints were assessed using modified intent-to-treat (mITT). The numerator was based on treated subjects who met the response criteria at follow-up Week 12; the denominator was based on all treated subjects.</td></tr>
<tr><td rowspan="3">Descriptive statistics and estimate variability</td><td>Treatment group</td><td>Cohort 1
Null Responder - Sentinel</td><td>Cohort 2
Null Responder - Expansion</td><td>Cohorts 3 & 4
IFN/RBV ineligible - naïve/intolerant expansion</td></tr>
<tr><td>Number of subjects</td><td>10</td><td>11</td><td>22</td></tr>
<tr><td>SVR12; Responder (%)</td><td>9/10 (90.0)*</td><td>10/11 (90.9)**</td><td>14/22 (63.6)***</td></tr>
<tr><td></td><td>80% CIs</td><td>(66.3, 99.0)</td><td>(69.0, 99.0)</td><td>(47.7, 77.5)</td></tr>
<tr><td>Notes</td><td colspan="4">* One subject failure: discontinued study drugs at Week 2 (HCV RNA > LLOQ at EOT); achieved SVR24 as documented in the follow-up SAE form provided by the investigator, but without follow-up Week 24 HCV RNA values in the clinical database.
** One subject failure: did not achieve < LLOQ, TD or TND at Week 4; added pegIFN
***Eight subject failures for SVR12 and SVR24: 3 with VBT; plus 1 discontinued at Week 8 (subject request), and was lost to follow-up post-treatment; 3 relapsed at follow-up Week 4; 1 relapsed at follow-up Week 12.</td></tr>
</table>

AI447017				
Analysis description	**Secondary analysis - Sustained Virologic Response at Follow-up Week 24 (HCV RNA < LLOQ, TD or TND at follow-up Week 24)**			
Analysis population and time point description	All analyses of secondary endpoints were based on all treated subjects (i.e., those in the sentinel or expanded cohort). In general, response rates for binary endpoints were assessed using modified intent-to-treat (mITT): the numerator was based on treated subjects who met the response criteria; the denominator was based on all treated subjects.			
Descriptive statistics and estimate variability	Treatment group	Cohort 1 Null Responder - Sentinel	Cohort 2 Null Responder - Expansion	Cohorts 3 & 4 IFN/RBV ineligible naïve/intolerant Expansion
	Number of subjects	10	11	22
	SVR24; Responder (%)	9/10 (90.0)*	10/11 (90.9)**	14/22 (63.6)***
	80% CIs	(66.3, 99.0)	(69.0, 99.0)	(47.7, 77.5)
Notes	* One subject failure: discontinued study drugs at Week 4 (HCV RNA > LLOQ at EOT); achieved SVR24 as documented in the follow-up SAE form provided by the investigator, but without follow-up Week 24 HCV RNA values in the clinical database.			
	** One subject failure: did not achieve < LLOQ, TD or TND at Week 4; added pegIFN			
	***Eight subject failures for SVR12 and SVR24: 3 with virologic breakthrough; plus 1 discontinued at Week 8 (subject request), and was lost to follow-up post-treatment; 3 relapsed at follow-up Week 4; 1 relapsed at follow-up Week 12.			
Analysis description	**Secondary analysis - Rapid Virologic Response at Treatment Week 4 (HCV RNA < LLOQ, TND at Week 4)**			
Descriptive statistics and estimate variability	Treatment group	Cohort 1 Null Responder - Sentinel	Cohort 2 Null Responder - Expansion	Cohorts 3 & 4 IFN/RBV ineligible - naïve/intolerant Expansion
	Number of subjects	10	11	22
	RVR; Responder (%)	4/10 (40.0)*	7/11(63.6)**	19/22 (86.4)***
	80% CIs	(18.8, 64.6)	(40.1, 83.1)	(72.1, 94.9)
Notes	* One subject discontinued study drugs at Week 2 (HCV RNA > LLOQ at EOT); achieved SVR24 as documented in the follow-up SAE form provided by the investigator, but without follow-up Week 24 HCV RNA values in the clinical database. Five additional subjects with HCV RNA data did not meet criteria for RVR and eRVR.			
	** One subject did not achieve < LLOQ, TD or TND at Week 4; added pegIFN after Week 6; failure (Week 4 futility rule) for all endpoints after Week 6. Three additional subjects had HCV RNA data (< LLOQ, TD) that did not meet criteria for RVR and eRVR.			
	*** Three subjects did not meet criteria for RVR.			

AI447017				
Analysis description	**Secondary analysis - Extended Rapid Virologic Response at Treatment Weeks 4 & 12 (HCV RNA < LLOQ, TND at Weeks 4 and 12)**			
Descriptive statistics and estimate variability	Treatment group	Cohort 1 Null Responder - Sentinel	Cohort 2 Null Responder - Expansion	Cohorts 3 & 4 IFN/RBV ineligible - naïve/intolerant Expansion
	Number of subjects	10	11	22
	eRVR; Responder (%)	4/10 (40.0)*	7/11 (63.6)**	17/22 (77.3)***
	80% CIs	(18.8, 64.6)	(40.1, 83.1)	(61.9, 88.5)
Notes	* One subject discontinued study drugs at Week 2 (HCV RNA > LLOQ at EOT); achieved SVR24 as documented in the follow-up SAE form provided by the investigator, but without follow-up Week 24 HCV RNA values in the clinical database. Five additional subjects with HCV RNA data did not meet criteria for RVR and eRVR.			
	** One subject did not achieve < LLOQ, TD or TND at Week 4 - added pegIFN after Week 6; failure (Week 4 futility rule) for all endpoints after Week 6. Three additional subjects had HCV RNA data (< LLOQ, TD) that did not meet criteria for RVR and eRVR.			
	*** Three subjects did not meet criteria for RVR, plus 1 subject had VBT at Week 10, and 1 subject discontinued at Week 8 with HCV RNA < LLOQ, TND and did not have an HCV RNA measurement at Week 12.			
Analysis description	**Secondary analysis - Virologic Failure**			
Descriptive statistics and estimate variability	Treatment group	Cohort 1 Null Responder - Sentinel	Cohort 2 Null Responder - Expansion	Cohorts 3 & 4 IFN/RBV ineligible - naïve/intolerant Expansion
	Number of subjects	10	11	22
	Virologic failure; n (%)	1 (10.0)*	1 (9.1) **	7 (31.8)***
Notes	* One subject in Cohort 1 (sentinel/prior null responder) who met the criteria for virologic failure on-treatment achieved SVR24 as documented in the follow-up SAE form provided by the investigator, but without follow-up Week 24 HCV RNA values captured in the database.			
	** 1 subject met the Week 4 futility rule			
	** 3 with VBT and 4 relapsed: In addition to the 7 subjects included in this table, 1 subject in Cohorts 3 & 4 discontinued study drugs at treatment Week 8, had HCV RNA < LLOQ, TND at Week 8, and was lost to follow-up.			

AI447017	
	www) Resistance analyses of the 7 failures meeting the requirement for resistance testing had resistance-associated substitutions to both investigational agents at or close to the time of virologic failure. xxx) The predominant NS5A amino acid substitutions were L31M/V-Y93H (7/7 failures), while NS3 protease amino acid substitutions were NS3-D168A (2/7 failures) and D168V (5/7 failures). yyy) All on-treatment failures carried the non-CC IL-28B GT (3/3 VBTs) while most relapsers (3/4) carried the CC IL-28B GT. **zzz)** The NS5A-Y93H resistance-associated polymorphism pre-existed in 23% (10/43) subjects and 50% (5/10) of subjects with this polymorphism subsequently failed treatment.

ASV - asunaprevir, BID - twice daily, BMS - Bristol-Myers Squibb, cEVR - complete early virologic response, CI(s) - confidence interval(s), DAA(s) - direct antiviral agent(s), DCV - daclatasvir, EOT - end of treatment, eRVR - extended rapid virologic response, GT(s) - genotype(s), HCV - hepatitis C virus, IFN - interferon, LLOQ - lower limit of quantification, mITT - modified intent-to-treat, pegIFNlo - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SVR - sustained virologic response, SVR24 - sustained virologic response at follow-up Week 24, TD - target detected, TND - target not detected

AI447011		
Title: Parallel, Open-label, Randomized, Multiple-dose Study to Evaluate the Safety, Pharmacokinetics and Pharmacodynamics of BMS-790052 and BMS-650032 in Combination in Null Responders to Standard of Care Infected with Chronic Hepatitis C Virus Genotype 1		
Study identifier	AI447011	
Design	Randomized, open-label, out-patient, multiple-dose, Phase 2a, pilot study with 2 parallel treatment groups and 2 parts: aaaa) **Part 1**: represented by the Sentinel Cohort (Treatment Groups A and B) with treatment duration up to 28 days and 2 study decisions at Weeks 2 and 4 bbbb) **Part 2**: represented by the duration after Week 4 of the Sentinel Cohort and the whole study duration of the Expansion Cohort. Expansion of a treatment group occurred only after the Sentinel Cohort satisfied criteria for successful response to treatment (SRT) at Week 2 and RVR at Week 4	
	Duration of main phase	**Part 1**: Subjects in the Sentinel Cohort were administered open-label DCV/ASV (Treatment Group A) or DCV/ASV/pegIFNα/RBV (Treatment Group B) for up to 28 days with 2 study decisions at Weeks 2 and 4. **Part 2**: cccc) Subjects in the Sentinel Cohort continued DCV/ASV (Treatment Group A) or DCV/ASV/pegIFNα/RBV (Treatment Group B) as long as all individual criteria for continuation were met. dddd) Subjects in the Expansion Cohort received open-label DCV/ASV (Expansion Cohorts A1 or A2) or DCV/ASV/pegIFNα/RBV (Expansion Cohorts B1 or B2) or DCV/ASV/RBV (Expansion Cohort B3) for up to 24 weeks.
	Duration of Follow-up phase	48 weeks post-treatment follow-up for the Sentinel and Expansion Cohorts (Parts 1 and 2)
Hypothesis	**Part 1:** The observed proportion of HCV GT-1 null responder subjects in the Sentinel Cohort with SRT is ≥ 70% at Week 2 and RVR is ≥ 50% at Week 4 for the combination of DCV/ASV with and without pegIFNα/RBV (SOC). Successful response to treatment was defined at Week 2 as either undetectable HCV RNA (< 10 IU/mL) or ≥ 2 $\log_{10}$ IU/mL decrease in plasma HCV RNA from baseline without rebound and at Week 4 by a RVR defined as undetectable HCV RNA (< 10 IU/mL). **Part 2:** The observed proportion of null responder subjects achieving SVR12 is ≥ 20%. SVR12 is defined as undetectable HCV RNA (< 10 IU/mL) at follow-up Week 12.	
Treatment groups	**Part 1:** 21 Sentinel subjects (GT-1a and -1b) were randomized (1:1) to Groups A or B	
	Group A - Sentinel	Subjects (GT-1a and -1b) were administered DCV 60 mg QD/ASV 600 mg BID Dual therapy for up to 24 weeks. ASV dose was reduced from 600 mg BID to 200 mg BID because of elevated transaminases noted in the Phase 2 study of ASV/pegIFNα/RBV (AI447016). At the time the ASV dose was lowered all subjects in the sentinel cohort of AI447011 had completed treatment with the exception of subjects with VBT, who were receiving rescue therapy (DCV/ASV/pegIFN/RBV). If rescue criteria were met, rescue therapy of DCV/ASV/pegIFNα/RBV was administered for up to 48 weeks.

AI447011			
	Group B - Sentinel	Sentinel subjects (GT-1a and -1b) received DCV 60 mg QD/ASV 600 mg BID/pegIFNα/RBV for up to 24 weeks. ASV dose was reduced from 600 mg BID to 200 mg BID because of elevated transaminases noted in the Phase 2 study of ASV/pegIFNα/RBV (AI447016). At the time the ASV dose was lowered all subjects in the sentinel cohort of AI447011 had completed treatment with the exception of subjects with VBT, who were receiving rescue therapy (DCV/ASV/pegIFN/RBV).	
	Part 2: Based on results from the Sentinel Cohort in Part 1, the decision was made to expand Treatment Groups A and B. An additional 38 subjects were randomized (1:1) into Expansion Cohorts A1 and A2. An additional 41 subjects were randomized (1:1) into Expansion Cohorts B1 and B2. Based on demonstration of adequate antiviral activity, an additional 22 subjects were enrolled into Expansion Cohort B3. Results for Group B3 were not included in the CSR.		
	Group A1 - Expansion	Subjects (GT-1b only) received DCV 60 mg QD/ASV 200 mg BID for up to 24 weeks. If rescue criteria were met, rescue therapy of DCV/ASV/pegIFNα/RBV was administered for up to 48 weeks.	
	Group A2 - Expansion	Subjects (GT-1b only) received DCV 60 mg QD/ASV 200 mg QD for up to 24 weeks. If rescue criteria were met, rescue therapy of DCV/ASV/pegIFNα/RBV was administered for up to 48 weeks.	
	Group B1 - Expansion	Subjects (stratified by GT-1a and -1b, and the total enrollment of GT-1b subjects was capped at 20% in each cohort) were administered DCV 60 mg QD/ASV 200 mg BID/pegIFNα/RBV for up to 24 weeks.	
	Group B2 - Expansion	Subjects (stratified by GT-1a and -1b, and targeted enrollment of GT-1b subjects < 20%) were administered DCV 60 mg QD/ASV 200 mg QD/pegIFNα/RBV for up to 24 weeks.	
	Group B3 - Expansion	Subjects (stratified by GT-1a and -1b, and targeted enrollment of GT-1b subjects < 20%) were administered DCV 60 mg QD/ASV 200 mg QD/RBV therapy for up to 24 weeks. If rescue criteria were met, rescue therapy of DCV/ASV/pegIFNα/RBV was administered for up to 48 weeks.	
Endpoints and definitions	Primary endpoints	Co-primary endpoints	**Part 1:** **Proportion of subjects with Successful Response to Treatment (SRT):** eeee) Proportion of subjects with either undetectable HCV RNA at Week 2 or ≥ 2 $\log_{10}$ IU/mL decrease in plasma HCV RNA from baseline without rebound during the first 2 weeks. Rebound was defined as ≥ 1 $\log_{10}$ IU/mL increase in HCV RNA from nadir either at more than 1 time point (not necessarily consecutive) or at last value through Week 2 or detectable RNA after achieving undetectable RNA. ffff) Proportion of subjects with RVR, defined as undetectable HCV RNA (< LLOQ-TND) at Week 4 **Part 2:** Proportion of subjects with SVR12, defined as undetectable HCV RNA (< LLOQ-TND) at follow-up Week 12

AI447011			
	Secondary endpoints	Secondary endpoint	**Part 1:** gggg) log_{10} HCV RNA change from baseline at Day 4, Day 7 and Day 14 **Part 2:** hhhh) Proportion of subjects with RVR, defined as undetectable HCV RNA at Week 4 on treatment iiii) Proportion of subjects with eRVR, defined as undetectable HCV RNA at both Weeks 4 and 12 on treatment jjjj) Proportion of subjects with cEVR, defined as undetectable HCV RNA at Week 12 on treatment kkkk) Proportion of subjects with SVR24, defined as undetectable HCV RNA at follow-up Week 24 llll) Frequency of genotypic substitutions associated with virologic failure
Database lock	03-Jan-2013		

Results and Analysis:

Part 1	**Part 1**		
Analysis description	**Primary Analysis: Successful Response to Treatment (SRT) at Weeks 2 and 4**		
Analysis population and time point description	These analyses were based on the sentinel cohort (Groups A and B) at Week 2 or Week 4. The proportion of subjects with antiviral activity endpoints was assessed using modified intent to treat (mITT); the numerator was based on treated subjects meeting the response criteria (regardless of add-on SOC); the denominator was based on all treated subjects. Response rates and 80% exact binomial CIs were presented by treatment group.		
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**	
		Group A DCV/ASV BID DUAL	Group B DCV/ASV BID/pegIFN/RBV QUAD
	Number of subjects	11	10
	Week 2 Successful Response* (Responder; %)	9/11 (81.8)**	9/10 (90.0)***
	80% CIs	(58.5, 95.1)	(66.3, 99.0)
Descriptive statistics and estimate variability	Treatment group	Group A Sentinel DCV/ASV	Group B Sentinel DCV/ASV/pegIFN/RBV
	Number of subjects	11	10
	RVR Week 4* (Responder; %)	7/11 (63.6)**	6/10 (60.0)***
	80% CIs	(40.1, 83.1)	(35.4, 81.2)
Notes	* SRT was defined as undetectable HCV RNA at Week 2 as either HCV RNA < LLOQ, TND (i.e., < 10 IU/mL) or ≥ 2 log_{10} IU/mL decrease in plasma HCV RNA from baseline without rebound and at Week 4 by a RVR defined as HCV RNA < LLOQ, TND.		

AI447011							
Analysis description	**Secondary Analysis: log_{10} HCV RNA Change from Baseline at Day 4, 7, and 14**						
Analysis population and time point description	This secondary antiviral activity endpoint analysis was based on the Sentinel Cohort (Groups A and B). The magnitude of the change in log_{10} HCV RNA (at Day 4, Day 7, and Day 14) was assessed by summarizing changes from baseline, including mean, standard deviation, 90% CIs, median and range by study day and treatment group.						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**					
		Group A DCV/ASV BID DUAL	Group B DCV/ASV BID/pegIFN/RBV QUAD				
	Number of subjects	11	10				
	Secondary Endpoint	**Mean log_{10} HCV RNA Change from Baseline to Day 4, 7, and 14**					
	Day 4 Mean (SD)	-4.2 (0.48)	-3.6 (0.50)				
	Day 7 Mean (SD)	-4.6 (0.40)	-4.1 (0.56)				
	Day 14 Mean (SD)	-5.3 (0.73)	-5.0 (0.80)				
Part 2	**Part 2:** Based on the antiviral activity results from the Sentinel Cohorts in Part 1, the decision was made to expand Treatment Groups A (A1 & A2) and B (B1, B2, & B3) and to continue with Part 2. Results for Group B3 are not presented in the CSR.						
Analysis description	**Primary Analysis: Sustained Virologic Response at Follow-up Week 12**						
Analysis population and time point description	The proportion of subjects with antiviral activity endpoints was assessed using mITT: the numerator was based on treated subjects meeting the response criteria; the denominator was based on all treated subjects. Response rates and 80% exact binomial CIs were presented by group.						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**		**Expansion Cohort**			
		DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	SVR12* Responder; (%)	4/11 (36.4)	10/10 (100)	14/18 (77.8)	13/20 (65.0)	19/20 (95.0)	20/21 (95.2)
	80% CIs	(16.9, 59.9)	(79.4, 100)	(60.4, 89.9)	(48.2, 79.3)	(81.9, 99.5)	(82.7, 99.5)
Note:	* Defined as HCV RNA < LLOQ, TND at follow-up Week 12.						
Analysis description	**Secondary Analysis: Rapid Virologic Response at Week 4**						
Analysis population and time point description	These secondary analyses were based on the Sentinel Cohort (A and B) and the Expansion Cohort (A1, A2, B1, and B2) at follow-up Week 12. In general, the proportion of subjects with antiviral activity endpoints was assessed using mITT: the numerator was based on treated subjects meeting the response criteria (regardless of add-on SOC); the denominator was based on all treated subjects. Response rates and 80% exact binomial CIs were presented by treatment group.						
Descriptive	Treatment	**Sentinel Cohort**		**Expansion Cohort**			

statistics and estimate variability	group	DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	RVR* Responder; (%)	7/11 (63.6)	6/10 (60.0)	12/18 (66.7)	11/20 (55.0)	15/20 (75.0)	15/21 (71.4)
	80% CIs	(40.1, 83.1)	(35.4, 81.2)	(48.8, 81.5)	(38.5, 70.7)	(58.5, 87.3)	(55.2, 84.2)
Notes	* Defined as undetectable HCV RNA < LLOQ, TND at Week 4						
Analysis description	**Secondary Analysis: Extended Rapid Virologic Response at Weeks 4 and 12**						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**		**Expansion Cohort**			
		DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	eRVR* Responder; (%)	4/11 (36.4)	6/10 (60.0)	11/18 (61.1)	10/20 (50.0)	14/20 (70.0)	15/21 (71.4)
	80% CIs	(16.9, 59.9)	(35.4, 81.2)	(43.3, 76.9)	(33.8, 66.2)	(53.3, 83.4)	(55.2, 84.2)
Notes	* Defined as undetectable HCV RNA <LLOQ, TND at both Weeks 4 and 12						
Analysis description	**Secondary Analysis: Complete Early Virologic Response at Week 12**						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**		**Expansion Cohort**			
		DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	cEVR* Responder; (%)	5/11 (45.5)	9/10 (90.0)	16/18 (88.9)	13/20 (65.0)	19/20 (95.0)	20/21 (95.2)
	80% CIs	(24.1, 68.2)	(66.3, 99.0)	(73.1, 97.0)	(48.2, 79.3)	(81.9, 99.5)	(82.7, 99.5)
Notes	* Defined as undetectable HCV RNA < LLOQ, TND at Week 12						

AI447011							
Analysis description	**Secondary Analysis: Sustained Virologic Response at Follow-up Week 24**						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**		**Expansion Cohort**			
		DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	SVR24* Responder; (%)	4/11 (36.4)	9/10 (90.0)	15/18 (83.3)	12/20 (60.0)	18/20 (90.0)	20/21 (95.2)
	80% CIs	(16.9, 59.9)	(66.3, 99.0)	(66.6, 93.7)	(43.3, 75.1)	(75.5, 97.3)	(82.7, 99.5)
Notes	* Defined as undetectable HCV RNA < LLOQ, TND at follow-up Week 24						

AI447011

Analysis description	**Secondary Analysis: Frequency of Genotypic Substitutions Associated with Virologic Failure**						
Descriptive statistics and estimate variability	Treatment group	**Sentinel Cohort**		**Expansion Cohort**			
		DUAL	QUAD	DUAL		QUAD	
		A ASV BID	B ASV BID	A1 ASV BID	A2 ASV QD	B1 ASV BID	B2 ASV QD
	Number of subjects	11	10	18	20	20	21
	VBT*	6/11	0/10	2/18	6/20	0/20	0/21
	Relapse	1/11	0/10	0/18	1/20	1/20	1/21

***Viral Breakthrough Definitions:**

Group A Sentinel Cohort

- Any increase in HCV viral load ≥ 1 log from nadir
- Any HCV RNA < LLOQ on or after Week 4
- Any HCV RNA < LLOQ, target detected (TD) on or after Week 4 confirmed by a subsequent consecutive HCV RNA measurement.

Expansion Groups A1, A2 and B3

- Any increase in viral load ≥ 1 log from nadir
- Any confirmed HCV RNA < LLOQ, TD on or after Week 8. Confirmation should have occurred via an immediate unscheduled return visit.
- Any HCV RNA ☐ L

Expansion Groups B1 and B2

- Any increase in HCV viral load ≥ 1 log from nadir
- Any confirmed HCV RNA [illegible] TND. Measurements were to be confirmed at the next scheduled visit.

****Viral Relapse Definition:** Viral relapse during the follow-up period was defined (in both Part 1 and Part 2) as confirmed HCV RNA ≥ LLOQ in a subject with HCV RNA < LLOQ, TD or TND at EOT.

A brief summary of the resistance results is provided below.

- HCV GT-1b prior null responders were less susceptible to virologic failure compared with GT-1a prior null responders when treated with Dual therapy.
- ASV dose impacted the virologic failure rate in HCV GT-1b prior null responders treated with Dual therapy; virologic failure was more common in subjects who received ASV 200 mg QD compared with ASV 200 mg BID.
- QUAD therapy (irrespective of ASV dose: ASV 600 mg BID, 200 mg BID, and 200 mg QD) was sufficient to suppress the emergence of resistance variants in subjects with GT-1a and GT-1b during therapy.
- In subjects treated with Dual therapy, the baseline (BL) nonstructural protein 5A (NS5A) resistance-associated polymorphism (RAP) tyrosine (Y)93 histidine (H) appeared to be associated with VBT in HCV GT-1b subjects.
- At the time of virologic failure (VBT or relapse), NS5A and nonstructural protein 3 (NS3) resistance variants were detected together. NS5A resistance variants included substitutions at glutamine (Q)30 (Q30 glutamic acid [E]/H/arginine [R]) and were often linked with other NS5A substitutions (leucine [L]31 methionine [M]/V, Y93H) in GT-1a and L31M/V-Y93H in GT-1b. NS3 resistance variants included R155K and aspartic acid (D) 168 alanine (A)/E/valine (V)/Y in GT-1a and D168V in GT-1b.

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	mmmm) In general, NS5A resistance variants persisted out to post-treatment Week 24 and beyond, irrespective of GT-1 subtype, whereas NS3 resistance variants were partially or completely replaced by baseline sequence. Emergent GT-1a NS3 resistance variants (R155K, D168E) appeared to be more fit than the predominant GT-1a and GT-1b D168V/Y variants.

ASV - asunaprevir, BID - twice daily, BMS - Bristol-Myers Squibb, cEVR - complete early virologic response, CI(s) - confidence interval(s), CSR - clinical study report, DCV - daclatasvir, GT(s) - genotype(s), eRVR - extended rapid virologic response, HCV - hepatitis C virus, LLOQ - lower limit of quantification, mITT - modified intent-to-treat, pegIFNα - peginterferon alfa, QD - once daily, RBV - ribavirin, RNA - ribonucleic acid, RVR - rapid virologic response, SNP(s) - single nucleotide polymorphism(s), SOC - standard of care, SRT - successful response to treatment, SVR - sustained virologic response, SVR12 - sustained virologic response at follow-up Week 12, SVR24 - sustained virologic response at follow-up Week 24, TND - target not detected, VBT - virologic breakthrough