# 2.0 Synopsis

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| **Abbott Laboratories** | **Individual Study Table Referring to Part of Dossier:** | **(For National Authority Use Only)** |
| **Name of Study Drug:** ABT-333 | **Volume:** | |
| **Name of Active Ingredient:** Sodium N-{6-[3-tert-Butyl-5-(2,4-dioxo-3,4-dihydropyrimidin-1(2H)-yl)-2-methoxyphenyl]naphthalen-2-yl}methanesulfonamide | **Page:** | |

**Title of Study:** A Follow-up Study to Assess the Evolution and Persistence of Resistance to ABT-333 After Discontinuation of ABT-333 Therapy in HCV Genotype 1-Infected Subjects Who Participated in Phase 1, 2, or 3 ABT-333 Clinical Studies

**Coordinating Investigator:** Maribel Rodriguez-Torres, MD

**Study Sites:** 7 sites in the United States and Puerto Rico.

**Publications:** There were no publications based on this study.

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| **Studied Period (Years):**<br>First Subject First Visit: 26 August 2008<br>Last Subject Last Visit: 27 May 2010 | **Phase of Development:** 2 |

**Objectives:** The objectives of this study were to assess the frequency and persistence of specific nonstructural protein 5B (NS5B) mutations selected by ABT-333 therapy, evaluate phenotypic resistance to ABT-333 in the in vitro subgenomic replicon system at serial time points, and correlate phenotypic resistance with NS5B mutations over time.

**Methodology:** This Phase 2 multicenter study was conducted in hepatitis C virus (HCV)-infected subjects who received ABT-333 at any dose level or matching placebo in Study M10-351 Substudy 2 and Study M10-380. After receiving at least 1 dose of ABT-333 or placebo, subjects were assessed for participation in this rollover study and asked to review the subject informed consent. The day of study completion or early discontinuation from the prior ABT-333 clinical study served as the baseline assessment for this rollover study. Subjects returned to the study site for their scheduled visits on an outpatient basis. Study visits were scheduled at Baseline and Weeks 2 and 4 and monthly through Week 48. Procedures at these visits included obtaining informed consent (at Baseline only), collecting 1 HCV RNA sample and 2 archive plasma samples, documenting any medications used for the treatment of HCV, and recording any serious adverse events that the investigator considered causally related to study procedures (i.e., venipuncture).

**Number of Subjects (Planned and Analyzed):** There was no planned number of subjects for this study because the number enrolled was to be based on the number of subjects who enrolled in other ABT-333 studies and subsequently agreed to participate in this study. Thirty-five subjects were enrolled in this study; 7 subjects were discontinued per protocol as the subjects had received placebo in the previous ABT-333 studies. Nine subjects had sufficient HCV RNA recovered from samples collected during this study (post-treatment with ABT-333 in the previous study) for genotypic and phenotypic analysis to proceed.

**Diagnosis and Main Criteria for Inclusion:**

Inclusion Criteria:

1. Subject received ABT-333 or matching placebo in a prior clinical study involving ABT-333.
2. Subject voluntarily signed and dated each informed consent, approved by an institutional review board, prior to the initiation of any study-specific procedures.
3. Subject was willing to commit to 12 months of study participation.

Exclusion Criteria:

1. The investigator considered the subject unsuitable for the study for any reasons inclusive of, but not limited to, failure to have complied with study procedures in a prior ABT-333 clinical study.

**Test Product, Dose/Strength/Concentration, Mode of Administration and Lot Number**: No study drug was dispensed in this study.

**Duration of Treatment:** No study drug treatment was dispensed in this study.

**Reference Therapy, Dose/Strength/Concentration and Mode of Administration and Lot Number:** No study drug was dispensed in this study.

**Criteria for Evaluation**

**Efficacy:** Efficacy was not assessed in this study.

**Resistance:** The prevalence of NS5B resistance mutations over time was assessed. The degree of phenotypic resistance (fold change in susceptibility compared to wild-type virus) to ABT-333 was also be assessed.

**Safety:** Only serious adverse events related to study procedures were assessed.

**Statistical Methods**

**Efficacy:** Efficacy was not assessed in this study.

**Resistance:** The prevalence of NS5B resistance mutations over time was summarized. If appropriate, the prevalence of mutations was summarized within subgroups of ABT 333-treated subjects defined by dose, duration of dosing, and/or concomitant therapy. The degree of phenotypic resistance (fold change in susceptibility to ABT-333 compared to baseline) was also summarized over time.

**Safety:** Serious adverse events were coded using the Medical Dictionary for Regulatory Activities and tabulated.

**Summary/Conclusions**

**Efficacy Results:** Efficacy was not assessed in this study.

**Pharmacokinetic Results:** Pharmacokinetics were not assessed in this study.

**Summary/Conclusions (Continued):**

**Resistance Results:** Five of the subjects showed no increase in concentration required for 50% effect ($EC_{50}$) relative to pretreatment $EC_{50}$. Four of these subjects ([redacted]) had received ABT-333 for 2 days, and had $EC_{50}$ values that were comparable to prototypic standard, both at pretreatment and post-treatment. A fifth subject ([redacted]) received ABT-333 600 mg BID for 28 days. This subject had an $EC_{50}$ value of approximately 30 nM both at pretreatment and at post-treatment, likely as a result of the Y448H variant which was present at pretreatment and maintained throughout the study. The remaining 4 subjects ([redacted]) received ABT-333 at doses from 300 mg BID to 1200 mg QD for 28 days, and had $EC_{50}$ values ranging from 3- to 33-fold higher in the post-treatment sample than at pretreatment. Subjects [redacted] were the only subjects that showed an increase in $EC_{50}$ post-treatment relative to baseline of greater than 5-fold. Both had relatively complex patterns of variants at multiple codons (5 and 15 codons, for Subjects [redacted], respectively). The only variant that they shared was at position 556. Subjects [redacted] had smaller increases in the frequency of 556G variants, and smaller increase in fold change $EC_{50}$. Fold increases in $EC_{50}$ for the four 556G-containing samples were 3-, 5-, 22- and 33-fold. For Subject [redacted], 36% of clones carried 556G 8 weeks after the last ABT-333 dose, but the increase in $EC_{50}$ relative to baseline was only 3-fold. Thus there is not a clear relationship between the prevalence of 556G and the level of resistance post-treatment, and there is potential for variants other than 556G to contribute to the resistance. However, the complexity of the pattern of variants does not permit attribution of resistance to specific variants other than 556G in these subjects.

Two types of comparisons of variants in the post-treatment sample were made: 1) comparison to the amount of that variant existing in the pretreatment sample; and 2) comparison of the change in the variant post-treatment relative to the amount of the variant present during the ABT-333 treatment (on-treatment) period. The number of variants per subject ranged from 1 to 15. The increase in the post-treatment samples in the percent of clones with each variant accumulated ranged from 9% to 87%. Twenty eight amino acid positions encoded variants that accounted for at least 10% more of the clones in the post-treatment sample than at pretreatment. For 23 of these positions, only 1 such instance occurred. At 2 positions (62 and 335), 2 of 8 subjects met the criteria for inclusion. Positions 117 and 543 showed variants in 3 of 8 subjects, and variants at position 556 were evident in 4 of 8 subjects. Of the 5 signature resistance variants previously identified for ABT-333, only the 556 variant was observed as enriched in the post-treatment samples.

Of note, in a majority of cases the frequency of variants observed in the post-treatment sample was either increased or unchanged relative to the on-treatment sample. In 5 instances, the number of variant clones decreased by 4% to 7% post-treatment relative to on-treatment. Eight of 37 instances showed little change (1% or less) in the percent of variant clones post-treatment relative to on-treatment. Sixteen instances showed an increase of 7% to 45%, with an additional 4 instances where the variants constituted 13% to 50% of total clones but the position did not meet the criteria for inclusion as an enriched variant in the on-treatment sample. Four of the 37 variants originated from a sample for which no on-treatment sample was tested. Of the positions with variants appearing in more than 1 subject, the only position that showed a trend in the change in accumulation relative to the on-treatment sample was 556, where the trend was towards increased accumulation of the glycine variant during the post-ABT-333 period. At each of the other 3 positions occurring in multiple subjects for which enrichment occurred on treatment (117, 335 and 543), there were both instances of increased frequency of variants from on-treatment to post-treatment samples, as well as instances where there was either no change or a decrease in frequency of variants.

**Summary/Conclusions (Continued):**

**Safety Results:** There were no serious adverse events related to study procedures during this study. No subjects died and no pregnancies were reported.

**Conclusions:** Nine subjects had sufficient HCV RNA recovered from samples collected during this study for genotypic and phenotypic analysis to proceed. One subject [redacted] did not have any new or enriched variants identified postbaseline. Five of the remaining 8 subjects were treated with ABT-333 for 28 days, while the other 3 were administered ABT-333 for 2 days. Thirty-seven instances of variants that were enriched relative to pretreatment prevalence were observed in these 8 subjects. These variants were distributed across approximately 5 percent of the NS5B codons, and most of the codons were only represented once in this group of subjects.

There were 23 codons at which enriched variants were detected in only 1 subject along with other enriched variants; thus in these instances a relationship between individual variants and phenotypic resistance could not be determined. For positions 62, 117, 335, and 543, which had variants detected postbaseline in either 2 or 3 subjects, there was at least 1 instance where there was no increase in $EC_{50}$ relative to pretreatment and 1 where there was a greater than 20-fold increase. All 4 subjects that were enriched in 556G showed an increase in $EC_{50}$ relative to pretreatment, ranging from 3- to 33-fold. This position was the only one of the signature ABT-333 resistance variants identified in previous studies to be present in the post-treatment samples, and each instance was associated with post-treatment resistance. The evidence from both this study and previous work indicates that 556G encodes resistance, and yet the prevalence of these variants increased after the treatment period for all of the subjects showing increased resistance post-treatment. This finding suggests that variants carrying 556G are sufficiently fit to persist and even expand in the absence of selective drug pressure, for at least as long as 44 weeks post-treatment in this study. Larger studies will be necessary to further quantify the durability of this mutation, as well as to assess the role of the other mutations detected in conjunction with 556G.

There were relatively few cases noted where a variant was enriched during treatment with ABT-333 and subsequently returned to pretreatment levels during the post-treatment period. Most of the post-treatment enriched variants either had become enriched during treatment and maintained (or increased) their prevalence during the post-treatment period, or had decreased in abundance during treatment and returned after treatment stopped. This analysis focused on variants that increased in quantity relative to pretreatment that might therefore have the potential to confer resistance to ABT-333 post-treatment. It is likely that a complete analysis of changes in variant prevalence would indicate that many of the changes noted were simply a function of the normal evolution of viral quasispecies, since many of these variants were already present at relatively high levels at baseline, prior to exposure to ABT-333.