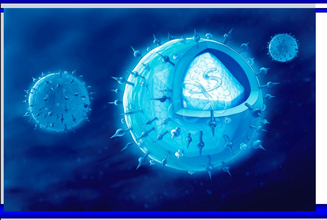

BIT225, AN HCV-P7 INHIBITOR, WITH ENHANCED ANTI-HCV ACTIVITY IN COMBINATION WITH PEGYLATED INTERFERON-ALFA-2B AND RIBAVIRIN IN HCV GENOTYPE-1 TREATMENT NAÏVE SUBJECTS: PHARMACOKINETIC AND RESISTANCE STUDIES

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INTRODUCTION

BIT225 is an inhibitor of the ion channel activity of Hepatitis C virus (HCV) protein p7, which is essential for the assembly and release of infectious HCV. The phase 2 trial reported here examines safety, pharmacokinetics and antiviral activity of BIT225 (28 days dosing) in combination with standard of care in treatment-naive subjects chronically infected with HCV Genotype 1.

STUDY DESIGN AND OBJECTIVES

The trial was a placebo-controlled, double-blind, randomized study designed to examine the safety and tolerability, pharmacokinetics (PK) and antiviral efficacy of two doses of BIT225 (200 and 400 mg), delivered orally twice a day for 28 days with standard of care (IFN/RBV*).

Twenty four treatment-naïve, genotype 1 subjects were enrolled from a single clinical site. After the 28 day BIT255 dosing phase all subjects remained on IFN/RBV for an additional 44 weeks.

[* *IFN/RBV: Pegylated-interferon alfa-2b (PEG-IFN) at doses of 80, 100, or 120 mcg weekly and ribavirin (RBV) 400 or 500 mg BID based on weight*.]

Primary objective :

• To evaluate the safety and tolerability of repeat dosing with BIT225

Secondary objectives :

• To evaluate the pharmacokinetic parameters of the BIT225 doses
• To evaluate the antiviral activity of BIT225+IFN/RBV compared to IFN/RBV alone

RESULTS

1. Baseline Characteristics

Baseline demographic and viral data were similar across the cohorts (data not shown).

The IL28B genotypes for 21 of 23 subjects were homozygous CC at 12979860 and homozygous TT at rs8099917. One subject from each of the 400mg BIT225 and placebo groups were heterozygous for both alleles.

2. Viral Load

At all time-points assessed; 4 weeks (RVR), 12 weeks (cEVR), and 48 weeks end of treatment (ETVR), the number of responders (HCV RNA < LLOQ) in the BIT225 cohorts were equal to or superior to the placebo group (Table 1). Most strikingly, after 48 weeks of therapy, the ETVR rate in the 400mg BIT225 group was 100%.

Table 1 – Summary of Viral Load Data

Cohort	RVR (Week 4)	cEVR (Week 12)	ETVR (Week48)
Placebo	2/8	3/8	6/8 (75%)
200 mg	2/8	5/8	7/8 (88%)
400 mg	2/7	4/7	7/7 (100%)

3. Pharmacokinetics

The PK of single doses of BIT225 (200 mg and 400 mg) was evaluated for each subject on day 0 and Day 28 with multiple samples collected up to 24 hours post-dose. Plasma concentrations of BIT225 were determined using a validated LCMS method and standard non-compartmental methods were used to estimate PK parameters (Phoenix 64, WinNolin 6.3). Pre-dose plasma concentrations were also measured on Days 3, 7, 10, 14 and 21.

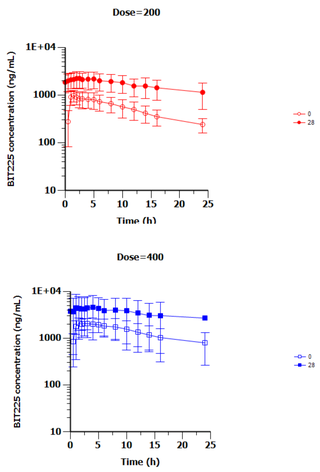

Table 2 – Summary of BIT225 Pharmacokinetic parameters on Day 0 and Day 28

	Day 0		Day 28	
Parameter [± 95% CI]*	200 mg (n=8)	400 mg (n=8)	200 mg (n=8)	400 mg (n=7)
C_{max}(ng/ml)	1092 [208]	2334 [512]	2356 [719]	4994 [3706]
T_{max} (hr) *Median*	2.38 [1.4] *1.50*	2.94 [1.3] *2.75*	2.95 [2.0] *2.03*	2.71 [1.7] *2.50*
AUC_{0-12} (ng/ml*hr)	8258 [2274]	20591 [6463]	23825 [7649]	48723 [35726]
AUC_{0-inf} (ng/ml*hr)	16934 [4382]	52356 [24912]	99594 [59246]	206651 [217748]
T1/2(hr)	13.2 [2.9]	15.1 [4.4]	32.2 [20.9]	21.6 [10.9]
Lambda-z(1/hr)	0.056 [0.011]	0.052 [0.018]	0.033 [0.018]	0.041 [0.020]
Vz/F(l)	355 [156]	289 [90]	272 [242]	194 [72]
Cl/F(l/hr)	18.5[7.1]	14.4[5.1]	5.75 [1.9]	8.84 [5.9]

Figure 1 – Mean (SD) plasma concentration versus time profiles: 200 mg group and 400 mg group, Day 0 and Day 28.

Summary of BIT225 pharmacokinetics

• Average C_{max} and AUC_{∞} values were consistent with a linear relationship of exposure for the two doses.
• Accumulation of BIT225 (~2-3 fold) occurred between the first dose and Day 3, after which time $C_{ss\text{-}min}$ trough concentration values were stable and consistent with elevated C_{max} and AUC_{∞} values seen on Day 28.
• A significant prolongation of terminal half-life (p=0.051) was observed between day 0 and day 28 for the 200 mg dose group, although statistical significance was not reached for the 400 mg dose, likely due to the large inter-individual variability.
• Consistent with increased half-life, oral clearance was reduced in both dose groups from day 0 to day 28. This suggests saturation of an elimination pathway, possibly due to a transporter or drug metabolism and warrants further investigation.

4. Safety and Tolerability

BIT225 was relatively well tolerated at both doses. The most common adverse events were fever, vomiting, headache, anemia, insomnia, and myalgia. One subject in the 400mg BIT225 cohort withdrew on Day 10 due to horizontal diplopia. Four other severe adverse events were recorded (2 of vomiting; 1 of vertigo and 1 of syncope) resulting in interruption of BIT225 treatment for periods of between 2 to 5 days early in dosing for three subjects. Symptoms resolved and all four subjects completed the trial. All other adverse events were of mild to moderate intensity.

5. E2-P7-NS2 Sequencing Analysis

The viral genes E2, P7 and NS2 were sequenced to determine if there were any alterations to the HCV population during antiviral therapy during the first 4 weeks of treatment. Several changes were identified but no unique mutations were selected in the BIT225 treatment cohorts compared to placebo. This suggests that there were no significant differences over the usual HCV quasispecies.

For the p7 protein sequence, there were 5 single site changes identified. (see Figure 2 below).

• No subject has more than one amino acid change in p7 between the paired samples.
• 4/5 of the substitutions were in subjects who had received either 400 or 200mg BIT225 (the other one was in a placebo recipient).
• One of the changes (L->M in S121) is in a highly conserved site and the 'M' is not seen in any of the other sequences.
• The change L->F in S124 is best considered a reversion to wild-type.
• The other 3 changes are observed in other isolates pre-BIT225 exposure.
• There was a relatively higher frequency of changes from baseline scattered throughout the E2 domain.

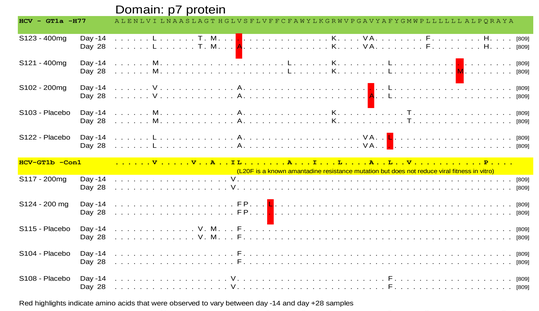

Figure 2 – Summary of genotypic changes in the P7 domain on Day 28 of genotype 1a and 1b participants treated with BIT225.

CONCLUSIONS

BIT225 is a first-in-class, specifically designed, HCV p7 inhibitor.

The PK data suggest that the dosing regimen generated sufficient drug levels in the plasma, resulting in promising antiviral activity. The data also suggests that there is the potential for less frequent dosing in future trials. Sequencing studies demonstrated that no BIT225-resistant variants were selected during therapy.

This study provides proof-of-concept of the validity of this approach to treatment of HCV infection in treatment-naïve genotype 1 subjects. Further clinical trials are in progress.

ACKNOWLEDGEMENTS

We would like to thank the University at Buffalo NY USA for the PK analysis, Southern Research Institute Fredrick MD USA for viral genomic sequencing and analysis, ACLIRES Inc for clinical trial assistance, the staff and trial participants at Sirijaj Hospital, Mahidol University, Bangkok, Thailand.

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