OBJECTIVES
To better understand imetelstat cellular distribution in tumor and bone marrow from xenograft mouse studies.
To establish a target tissue pharmacokinetic model to simulate imetelstat human tumor and bone marrow concentration profiles.

METHODOLOGY

RESULTS AND DISCUSSION

INTRODUCTION
Imetelstat is a first-in-class, illustrated 15-oxoroligocarbonate \( \text{Rho}-\)phosphorhamidate telomerase inhibitor with anti-tumor activity. It has a sequence that is complementary to the hTR template region in telomerase. It inhibits telomerase by acting as an active site competitive inhibitor of the enzyme in the sub-nanomolar and sub-micromolar range in biochemical and cellular assays, respectively.1 Imetelstat has demonstrated telomerase inhibitory and cancer growth inhibitory effects in both in vitro and in vivo preclinical models.2-5 Imetelstat is currently in Phase II clinical development. Its distribution in tissues, specifically tumor and bone marrow, was studied in a xenograft mouse model established using an hOCAR-5 cell line. Data from xenograft studies indicated imetelstat tumor and bone marrow concentrations were sustained throughout the terminal phase. This suggests that further evaluation of imetelstat distribution in patient tumor and bone marrow is critical for understanding the determinants for drug tissue accumulation in order to facilitate clinical understanding of drug concentration in the tissue of the hematologic and solid tumor malignancies.

Human tumor and bone marrow sample are difficult to obtain. It is essential to assay serial human tissue concentrations as a function of time. Single time point tumor or bone marrow samples may be obtained through biopsy or surgical procedures. The timing of the samples are difficult to control. Therefore, the purpose of this study is to develop a target tissue pharmacokinetic model, to predict overall imetelstat human tumor or bone marrow concentration profiles from a single tissue sample at a random time point. Therefore, we provide a prediction for overall tumor-bone marrow drug exposure in patients. PK data from preclinical xenograft mouse studies and modeling are presented.

Data from Xenograft Mouse Study

- Imetelstat concentrations in plasma, tumor, and bone marrow were quantified in xenograft mouse samples. For simplicity, detailed concentration-time profiles are only shown for 30 and 90 mg/kg dose groups in Figure 1 and 2.

- Plasma samples were flash frozen immediately after collection, and were homogenized in M-Per buffer with Omni hand homogenizer (Kerexen, CT). Bone marrow samples were collected from both femurs by FBS flushing, and bone marrow cells were lysed in M-Per buffer.

- All samples were subjected to a novel extraction procedure using a bistynlated capture probe before imetelstat concentration determination with on-painting reversed-phase LC/MS/MS. Imetelstat concentration in bone marrow was presented as total concentration, which accounts for both the cellular and non-cellular fractions.

Target tissue pharmacokinetic model: Data from preclinical xenograft mouse studies were used in developing a pharmacokinetic model to describe imetelstat tissue disposition in tumor and bone marrow. All programs were written in WinNonlin (Pharsight Corp) model.

- Human plasma from solid tumor (ET) patients. Human plasma data was obtained from solid tumor patients in a Phase I clinical trial. The 2 h or 4 h intails of imetelstat at 9.4 mg/kg. Imetelstat plasma concentration were determined by a LC/MS-based assay.

- Human bone marrow from multiple myeloma (MM) patients. Imetelstat concentrations in tumor and bone marrow were at higher levels than that in plasma when equilibrium was established after 24 hrs following distribution between tissue and plasma. Imetelstat tissue degradation in the first 24 hrs, and maintains a constant ratio only after equilibrium is achieved during the terminal phase. This was observed in all dose groups, and is in agreement with literature reports on other stigmastanoids.6 The absolute concentration ratio between tissue and plasma in the terminal phase varies among patients. Therefore, no saturation in exposure is observed in tumor and bone marrow at the highest dose of 90 mg/kg.

Data from Xenograft Mouse Study

- Imetelstat concentration-time profile following continuous IV infusion was similarly modeled with only the adjustment of the input function. The rate constant from central to tissue compartment, k9, was assumed to be similar between human and xenograft mouse. Using this model, together with human plasma data at 9.4 mg/kg, imetelstat tissue concentrations in human tumor and bone marrow were simulated as shown in Figure 4.

- Bone marrow concentration-time profile was predicted for a NKI Phase I patient, based on the observed plasma data (Figure 5). The predicted imetelstat concentration at 24 hrs after dosing is 0.4 mg/g in bone marrow in concordance with the observed value of 5.5 ug/g.

Target Tissue PK Modeling

- Xenograft datasets were used in building a preclinical pharmacokinetic model to describe imetelstat tissue distribution in tumor and bone marrow. Imetelstat concentration-time profiles in plasma and tissue were modeled simultaneously. Plasma concentrations followed a biphasic decline, and were modeled as a two compartment open model with a first order input. Tissue compartments (effect compartment, tumor or bone marrow) were modeled as a linked compartment from the central compartment. Imetelstat transfers from the central to the tissue compartment via a rate constant, k6 (Scheme 5). Simulated plasma- and tissue-level concentration profiles in xenograft mouse can be seen in Figure 3.

- Imetelstat distribution rapidly into xenograft tumor and bone marrow. Imetelstat concentrations in tumor and bone marrow were at higher levels than that in plasma when equilibrium was established after 24 hrs following distribution between tissue and plasma. Imetelstat tissue degradation in the first 24 hrs, and maintains a constant ratio only after equilibrium is achieved during the terminal phase. This was observed in all dose groups, and is in agreement with literature reports on other stigmastanoids.6 The absolute concentration ratio between tissue and plasma in the terminal phase varies among patients. Therefore, no saturation in exposure is observed in tumor and bone marrow at the highest dose of 90 mg/kg.

- Human tumor concentration-time profile following continuous IV infusion was similarly modeled with only the adjustment of the input function. The rate constant from central to tissue compartment, k9, was assumed to be similar between human and xenograft mouse. Using this model, together with human plasma data at 9.4 mg/kg, imetelstat tissue concentrations in human tumor and bone marrow were simulated as shown in Figure 4.

CONCLUSIONS

1. A target tissue pharmacokinetic model was established to describe imetelstat tumor and bone marrow concentration profiles in xenograft mice.
2. The current model will serve as a starting point for human target tissue prediction. It will allow the estimation of maximum target tissue concentration (Cmax) and overall target tissue exposure (AUC) from sparse clinical sampling.
3. Once optimized, a variety of preclinical and clinical tissue measurements, the model may be used to predict final target tissue PK, and guide clinical trial dosing and dosing regimens.
4. Single patient bone marrow data was in good concordance with the model.
5. Additional patient plasma, tumor and bone marrow data will be valuable for refinement of the target tissue PK model. Human tumor has greater variability than preclinical xenograft models. Therefore, tumor variability will need to be assessed by sampled validated patient samples from the clinical setting.
6. The models application to other tumor types (e.g. primary brain tumor) will be assessed from patient samples derived from future studies.

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References