Characterizing the PK/PD relationship of C/EBPβ antagonist ST101 in a mouse orthotopic breast cancer model

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Introduction

Sapience Therapeutics is focused on discovering and developing peptide-based therapeutics to previously 'undruggable' targets for major unmet medical needs, particularly high mortality cancers.

ST101 is a peptide antagonist of the oncogenic transcription factor CCAAT/Enhancer Binding Protein Beta (C/EBPβ). ST101 is currently being evaluated in a Phase 1/2 clinical study in patients with advanced unresectable and metastatic solid tumors, with expansion cohorts in cutaneous melanoma, glioblastoma, hormone receptor positive breast cancer and castrate-resistant prostate cancer.

Abstract

Transcription factor dysregulation is common in cancer, resulting in gene transactivation that drives oncogenesis. The transcription factor C/EBPβ is aberrantly activated in many cancers, where it drives a genetic profile that promotes survival and proliferation of tumor cells and inhibits differentiation. ST101 is a peptide antagonist of C/EBPβ that prevents its dimerization with oncogenic cofactors by binding the leucine zipper domain, ultimately targeting C/EBPβ for proteasomal degradation. Here we describe the pharmacokinetic/pharmacodynamic (PK/PD) profile of ST101 in a non-clinical orthotopic breast cancer model. An integrated population pharmacokinetic/pharmacodynamic (PK/PD) model was developed to describe the exposure−response relationship for ST101 anti-tumor activity in female Balb/c mice bearing 4T1-luc triple-negative breast cancer (TNBC) orthotopic tumors. Exposure−response predictions were made based on estimated potency from the PK/PD model and mean plasma concentrations observed at the doses investigated. Approximately 100% tumor growth inhibition (TGI) was observed at the highest doses administered in this study. The AUC values required to achieve IC_{50} and IC_{90} were 80,500 and 99,600 ng/ml*hr, respectively. Weekly cumulative doses in mice required to achieve 50% and 90% TGI were estimated as approximately 97 mg/kg and 120 mg/kg per week. The integrated model provides a useful tool to characterize and predict the ST101 dose-response relationship, provide a quantitative rationale for dose selection in a phase 2 clinical trial and supports the continued development of ST101 as a potent peptide therapeutic for patients with solid tumors.

ST101 Mechanism of Action

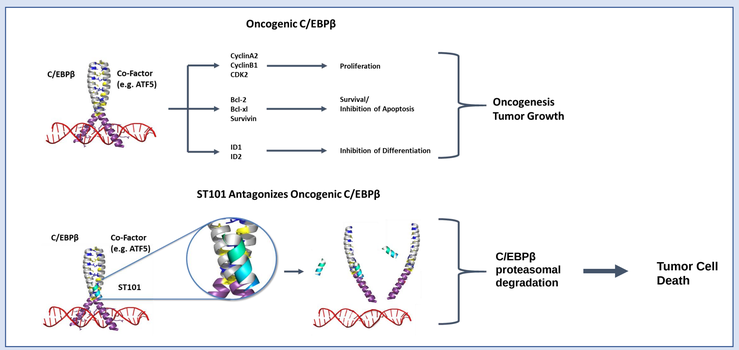

Figure 1: ST101 mechanism of action. C/EBPβ overactivation in breast cancer drives tumor cell proliferation, survival and inhibits differentiation. ST101 disrupts C/EBPβ dimerization with oncogenic cofactors, preventing C/EBPβ mediated transcription and enhancing proteasomal degradation. The result is antagonism of oncogenic gene transactivation leading to selective tumor cell death.

Results

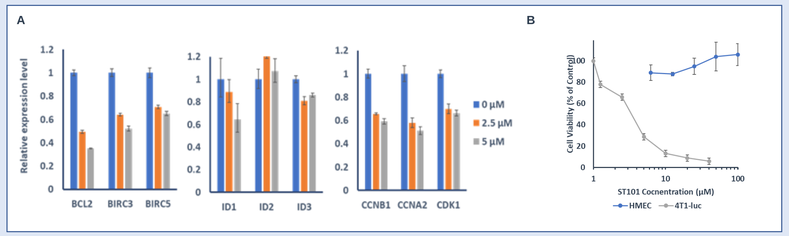

Figure 2: ST101 Antagonizes C/EBPβ Target Gene Expression and Induces Cell Death in TNBC Cells. A) qPCR analysis of RNA extracted from 4T1-luc TNBC cells following 24 hr exposure to ST101 (0, 2.5, or 5 µM). Expression of genes involved in survival (BCL2, BIRC3, BIRC5), differentiation (ID1, ID2 and ID3) and cell cycle (cyclin B1, cyclin A2 and cyclin-dependent kinase 1) were quantified. Data represents log2 normalized expression (2^ΔΔCt) and standard error of mean. B) Cell viability of 4T1-luc TNBC or normal human mammary epithelial cells (HMECs) following exposure to ST101 for 48 hrs. Viability was analyzed following annexin V/PI staining by high-content imaging and is presented as percent of vehicle-treated control. Data represents mean±SEM for a minimum of 3 replicates for each data point.

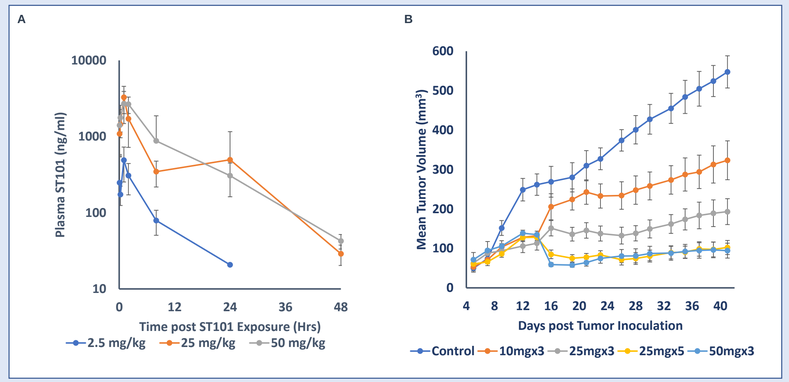

Figure 3: ST101 Displays Anti-Breast Cancer Activity in vivo. A) Female C57BL/6 mice were administered a single dose of ST101 by subcutaneous injection. Plasma exposure levels were quantified by LC-MS. B) Female balb/c mice were injected with 1x10^6 4T1-luc TNBC cells in the 4th mammary fat pad. ST101 (10, 25 or 50 mg/kg) was administered 3x/week, or 25 mg/kg was administered 5x/week, for the duration of the study, resulting in 45.6%, 73.8% and 95.4% and 95.1% tumor growth inhibition (TGI), respectively. Wilcoxon matched-pairs signed rank test indicates significant difference in tumor volume following 25 and 50 mg/kg ST101 vs vehicle control ($p<0.01$ and $p<0.001$, respectively; n=6 mice/group). No significant impact of ST101 on mouse body weight was observed during the study.

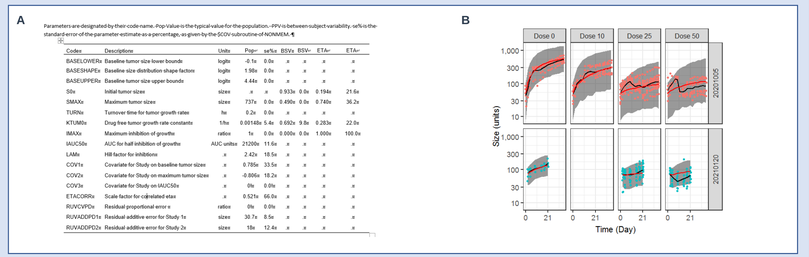

Figure 4: A tumor growth PK/PD model was built to characterize ST101 in vivo activity. A) A model was initially developed for drug-free tumor growth. Tumor growth was defined by an underlying rate constant for tumor growth (KTUM) using a turnover model that dictated the growth rate to maximum tumor size. A logistic style function constrained baseline tumor size. Drug affected KTUM via a sigmoid inhibitory relationship. Maximum inhibition (IMAX) was fixed to 0 (complete inhibition). Two studies were included with different baseline and maximum tumor size, which was accounted for using covariates. B) Visual predictive check plot of the model fit. The dots are the observed values (pink indicates study 1 and cyan indicates study 2). The black line is the median observed values; the red line is the median of simulated tumor sizes; the grey ribbon shows the 90% prediction intervals for the simulated data. Overall, the observed values are well-predicted based on these plots.

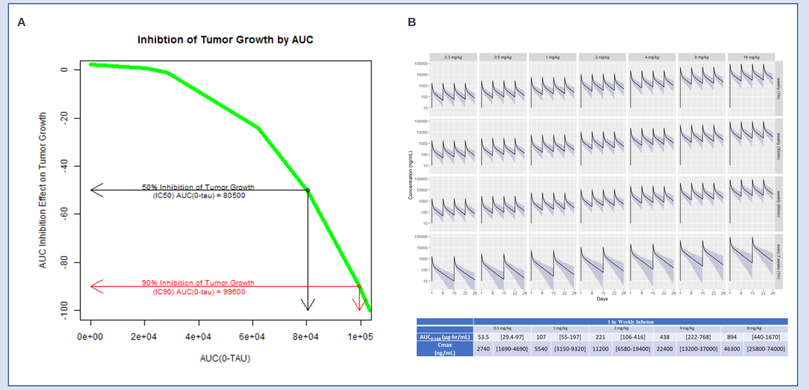

Figure 5: PK/PD model indicates that AUC predicts ST101 anti-tumor activity. A) Increasing AUC values result in increased tumor growth inhibition. The AUC value that results in 50% (IC_{50}) and 90% (IC_{90}) TGI are 80,500 and 99,600 ng/ml*hr, respectively. B) Population PK (PPK) modeling from preclinical cynomolgus monkey studies was used to predict ST101 concentrations in a 70 kg human at various dose levels following a 1-hour intravenous infusion. The PPK of ST101 is characterized by a 2-compartment disposition model with zero-order (intravenous) input and first order elimination. The model utilized traditional allometric scales, i.e., 0.75 for CL and Q, 1 for Vc and Vp. Human body weight (70kg) was normalized by approximate monkey body weight (3 kg). A once weekly 2 mg/kg ST101 infusion is predicted to achieve an $AUC_{0\text{-}168}$ that exceed the IC_{90} in mouse tumor models. The table summarizes AUC (µg/ml*hr) and C_{max} ((µg/m) from the simulated ST101 dose levels.

Conclusions

- Significant tumor growth inhibition (TGI) was observed following ST101 administration in 4T1-luc TNBC orthotopic tumor model ($p<0.001$ for 50 mg/kg ST101 vs. control).
- The AUC values associated with the ST101 IC_{50} and IC_{90} are 80,500 and 99,600 ng/ml*hr, respectively. The dosing regimen in mice required to achieve IC_{50} and IC_{90} are estimated as approximately 97 mg/kg and 120 mg/kg per week, respectively.
- PPK modeling from preclinical studies (cynomolgus monkeys) predicts that 2 mg/kg ST101 infusion in humans will exceed exposures associated with the IC_{90} in mice.
- In the ongoing Phase 2 dose expansion part of the study, Sapience has initiated dosing in patients with GBM and metastatic cutaneous melanoma, and will soon commence dosing in patients with refractory, locally advanced or metastatic hormone-receptor-positive breast cancer and castrate-resistant prostate cancer.