Abst. No. 2947

Pharmacodynamic (PD) and Pharmacokinetic (PK) Results of the Second-generation Hypomethylating Agent, SGI-110, in Patients with Hepatocellular Carcinoma (HCC) after Progression on Sorafenib

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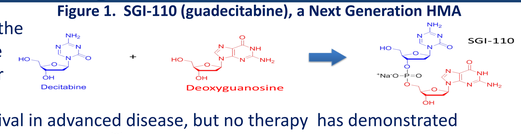
Background

- Hepatocellular Carcinoma (HCC) is the sixth most common cancer and the third most common cause of cancer death worldwide [(1)]
- Sorafenib treatment improves survival in advanced disease, but no therapy has demonstrated significant activity after progression on sorafenib [(2)]
- SGI-110, a dinucleotide of decitabine and deoxyguanosine (Fig 1), affords increased in vivo exposure of decitabine by protecting it from deamination due to slow release upon SQ injection
- In Phase 1 AML/MDS studies, SGI-110 provides longer exposure and more potent hypomethylation compared to decitabine [(3)]
- Preclinical studies demonstrated:
  - In vitro, SGI-110 induced significant hypomethylation of tumor suppressor genes RASSF1A, SOCS1 and DAB2IP in human HCC cell lines HuH7 an HepG2 and resulted in a potent reduction in colony formation at low nanomolar concentrations of SGI-110 [(4)]
  - SGI-110 efficiently sensitizes HCC cells and xenografts to oxaliplatin by inhibiting distinct signaling pathways, allowing for high antitumor activity without systemic toxicity (Kuang et al., AACR 2015, Abst 2533)
  - Numerous epigenetic alterations accumulate during hepatocarcinogenesis, leading to activation of oncogenes or loss of tumor suppressor genes in HCC. Specifically, increased methylation of genes implicated in HCC tumorigenesis has been associated with pathogenesis and poor outcome
- In this study, we evaluated therapeutic and biologic effects of SGI-110, a hypomethylating agent (HMA)in patients with HCC. PK and PD results of this open-label, phase 2 study in patients with HCC are presented here

Figure 1. SGI-110 (guadecitabine), a Next Generation HMA



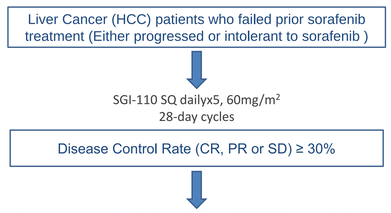
STUDY DESIGN

This is an open-label, single-arm, non-randomized Phase 2 study using Simon's 2-Stage Design :

- Stage 1: A minimum of 15 evaluable subjects will be enrolled and at least 6 subjects need to demonstrate disease control at 16 weeks to continue
- Stage 2: Once disease control is demonstrated, an additional 31 subjects will be enrolled

(Disease Control Rate (DCR)- Is defined as percentage of subjects who achieve a best clinical response of CR or PR plus subjects who are SD at 16 weeks using RECIST v1.1)

Figure 2. Study Design



Major Study Objectives

- To assess the Disease Control Rate (DCR) at 16 weeks for subjects treated with SGI-110 after failure of sorafenib
- To assess the safety and tolerability of SGI-110 treatment
- To determine progression-free survival (PFS) and overall survival (OS) of SGI-110 treatment
- To determine alpha fetoprotein (AFP) response to SGI-110 treatment
- To determine the biological effect of SGI-110 on methylation of long interspersed nucleotide elements-1 (LINE-1) in blood and tumor tissue
- To determine the methylation status, and re-expression of silenced genes, in particular tumor suppressor genes (TSGs) such as RASSF1A, SFRP, MZB1, P16, and others, after SGI-110 treatment

Major Inclusion and Exclusion Criteria

Inclusion

- Subjects 18 years of age or older, ECOG 0-1, acceptable organ function, and life expectancy of at least 16 weeks
- Histologically or cytological confirmed advanced stage HCC and failed prior sorafenib treatment defined as demonstrating progressive disease OR sorafenib intolerance despite at least one dose reduction

Exclusion

- Subjects with known hypersensitivity to SGI-110, poor medical risk due to systemic diseases, active uncontrolled infections, or life threatening illness that could compromise subjects safety or interfere with absorption or metabolism of SGI-110
- Subjects with, uncontrolled Ischemic heart disease or h/o congestive cardiac failure >= grade 3 per NYHA Subjects with the following conditions associated with cirrhosis or HCC:
  - Known brain metastases or active hepatitis B infection
  - Encephalopathy or variceal bleeding (esophageal or gastric) within the last 6 months
  - Subjects with clinically evident ascites
  - Child-Pugh C cirrhosis or Child-Pugh B cirrhosis with more than 7 points
- Subjects with prior malignancy, known history of human immunodeficiency virus (HIV)

Study Conduct

- Dosing in 28 day cycles:
  - SGI-110 daily days 1-5 of a 28-day cycle
    - Initial dose 60mg/m^2
    - Due to myelosuppression seen in first 4 subjects , the dose was reduced from 60 mg/m^2 to 45 mg/m^2 for subsequent patients
- PK/PD samples collected pre and post dosing
- Disease assessment (radiological): every 8 weeks
- Subjects are encouraged to remain on treatment for at least 6 cycles because delayed responses have been observed with HMAs
- Pharmacodynamic Samples:
  - Tumor Biopsies at screening and C1D8 or C2D8: Global (LINE-1) and gene specific DNA methylation
  - Blood at C1D1 and C1D8 : Global (LINE-1) DNA methylation

Figure 3. Schema

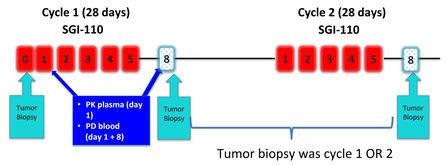


Table 1. Patient Characteristics and Dosing Cohorts (SGI-110)

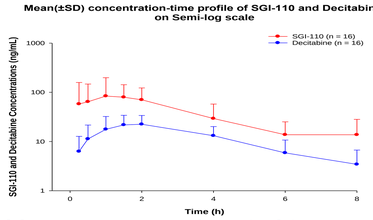
Characteristics	60 mg/m^2 (n=4)	45 mg/m^2 (n=46)	Total=50
Age (mean, range)	52 (48-57)	61 (32-82)	60(32-82)
ECOG			
0	1 (25%)	20 (43%)	21(42%)
1	3 (75%)	26 (57%)	29(58%)
Child Pugh Score			
Class A	4 (100%)	38(83%)	42(84%)
Class B	0	8(17%)	8(16%)
Scores (mean, range)	5.8(5-6)	5.7(5-7)	5.7(5-7)
Number of prior regimens (median, range)	2.5(1-3)	2(1-8)	2(1-8)

Safety

Table 2. Summary of all Related Grade 3 and Above Adverse Events to SGI-110

	60mg/m^2 (n=4)		45 mg/m^2 (n=46)		Total	
Events	# and % of Subjects		# and % of Subjects		# and % of Subjects	
Neutropenia	4	100%	31	67.4%	35	70%
Leukopenia	1	25%	13	28.3%	14	28%
Febrile Neutropenia	1	25%	5	10.9%	6	12%
Aspartate aminotransferase increased	1	25%	4	8.7%	5	10%
Thrombocytopenia	4	100%	1	2.2%	5	10%
Lymphopenia	0	0	5	10.9%	5	10%
Anaemia	1	25%	1	2.2%	2	4%

Figure 4. SGI-110 and Decitabine Concentration-time Profile



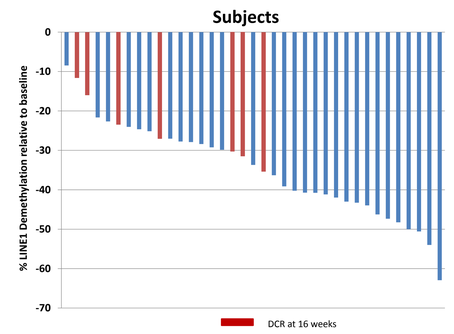
- PK profiles (Figure 4) for SGI-110 and decitabine show longer presence of both analytes than in study 01 (AML/MDS)

Table 3. SGI-110 and Decitabine PK Parameters

Pharmacokinetic parameters				
SGI-110				
PK Parameters	N	Mean	SD	CV%
$AUC_{0\text{-last}}$ (ng*hr/mL)	16	288	190	65.8
C_{max} (ng/mL)	16	106	107	100
Decitabine				
$AUC_{0\text{-last}}$ (ng*hr/mL)	16	93.9	22.1	23.5
C_{max} (ng/mL)	16	28.0	11.5	41.2

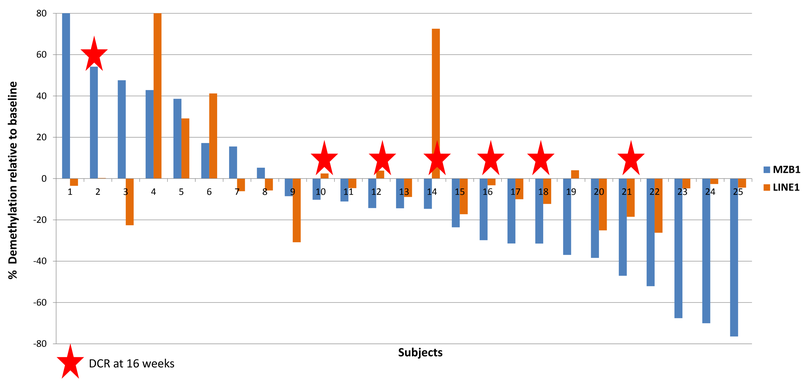
- Serial blood samples were collected to derive plasma on C1D1 from 0-8 hours after SGI-110 administration from 16 subjects. SGI-110 and decitabine levels in plasma were determined using a validated LC-MS/MS method with LLOQ of 1 and 0.5 ng/mL, respectively
- Pharmacokinetic parameters were estimated and descriptive statistics derived by non-compartmental analysis using Phoenix WinNonlin® (v.6.1)
- Mean $AUC_{0\text{-last}}$ for active metabolite decitabine in this study after the SGI-110 dose of 45 mg/m^2 was 93.9 ng*hr/mL which was higher than projected from 01 based on doses of 36 and 60 mg/m^2

Figure 5. LINE-1 Demethylation in Peripheral Blood



- Overall the mean LINE-1 demethylation relative to baseline was – 34.5% ± 12.06
- Demethylation was observed **in blood** from all 37 subjects, including 7 subjects with Disease Control at 16 weeks

Figure 6. LINE1 & MZB1 Demethylation for Individual Tumor Biopsies



- **LINE-1 was analyzed in 32 paired tumor biopsies (data shown for 25 subjects with both markers)**
  - LINE-1 demethylation in tumor DNA was observed in 22 subjects (68.7%).
  - Overall average demethylation was -14.2%
  - Among these 22 subjects, biopsies obtained C2D8 (n=18) had an average demethylation of -15.9% which was higher than the demethylation observed in biopsies obtained C1D8 (n=4) of -6.7%
- MZB1 is a recently identified tumor suppressor gene frequently hypermethylated in HCC [(5)]
- Methylation-mediated silencing of MZB1 (marginal zone B and B1 cell-specific protein) is known to lead to loss of its tumor-suppressive activity
  - Demethylation of MZB1 reduces the cell proliferation of HCC cells
- **Paired tumor biopsies were obtained for MZB1 demethylation analysis from 25 subjects.**
  - MZB1 demethylation in tumor DNA was observed in 17 subjects (68%)
  - Overall average demethylation was -34%
  - Six out of 7 subjects with disease control at 16 weeks showed potent MZB1-specific demethylation

Conclusions/Summary

- SGI-110 dosed at 45mg/m2 D1-5 on a 28-day cycle was generally well tolerated in an HCC population previously progressed on sorafenib
- Potent global DNA demethylation (LINE-1) was observed in blood and tumor DNA
- Extent of LINE-1 demethylation apparently was higher on cycle 2 than cycle 1 in tumor biopsies
- Demethylation was also observed in a majority of subjects on promoter of tumor suppressor gene MZB1 which is frequently hypermethylated and silenced in HCC
- Changes in blood and tumor LINE-1 and MZB-1 methylation are promising and consistent with the desired biologic effect of SGI-110
- Comparison of data with previously performed studies (study 01 AML+MDS) suggests that both SGI-110 and decitabine exposures in HCC subjects appear to be higher and more persistent[(3)]

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Poster presented at: AACR Annual Meeting, Philadelphia, PA April 18 - 22, 2015

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