Preliminary results of a Phase I study of SZN-043, a Novel R-Spondin mimetic, in Healthy Volunteers and Subjects with Liver Cirrhosis

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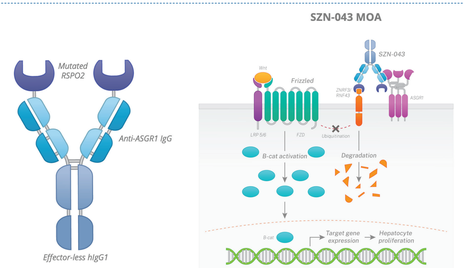
Introduction

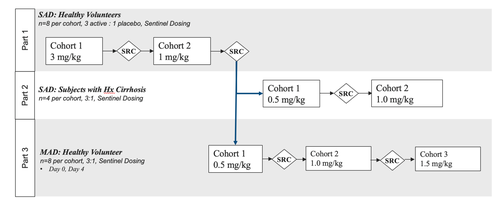
- Severe alcohol-associated hepatitis (SAH) is a medical area of high unmet need, with no current approved medications. As the only guideline recommended medication, corticosteroids have not been associated with improved long-term survival benefit. Retrospective research has found no improvement in survival with medical management during the last 60 years. SAH is associated with impaired hepatocyte proliferation.
- Elevated Wnt signaling and increased hepatocyte proliferation have been linked to greater survival, suggesting that therapies that can enhance hepatocyte proliferation can benefit patients. R-spondins (RSPOs) are known enhancers of Wnt signaling. SZN-043 is a bispecific fusion protein and hepatocyte-specific RSPO mimetic shown to induce hepatocyte-targeted Wnt signaling and hepatocyte proliferation in preclinical studies (Figure 1).
- SZN-043 was evaluated for safety, pharmacokinetics, and pharmacodynamics in a single center, first-in-human, Phase 1, randomized, double-blind, placebo-controlled, single ascending dose (SAD) and multiple ascending dose (MAD) study in healthy volunteers (HV) and patients with a history of liver cirrhosis (PHLC).

Figure 1. Structure and mechanism of action of SZN-043



Methods

Figure 2. Design of Phase Ia study



- Subjects enrolled in Parts 1, 3 were between the ages of 18–50 years of age found to be in good health
- Subjects enrolled in Part 2 were patients with a history of cirrhosis (PHLC) with a Child-Pugh score between 5 and 7, inclusive; a fibroscan of ≥ 6; and a MELD score ≤ 12
- SZN-043 was administered as an IV infusion on Day 0 (Parts 1, 2) or Days 0, 4 (Part 3)
- Subjects were followed for 4 weeks after their last dose
- Safety and tolerability assessed according to the following: Vital signs, physical examinations, clinical laboratory tests (hematology, coagulation, serum chemistry, CRP [c-reactive protein], GLDH [glutamate dehydrogenase], CK18, complement, cytokine panel, PBMC [peripheral blood mononuclear cell], and urinalysis), ECGs, and review of TEAEs and TESAEs should they occur.
- Pharmacokinetic evaluation was conducted based on serum measurements following dosing
- Pharmacodynamic evaluation included occupancy of ASGR1 as measured by serum alkaline phophatase, methacetin metabolism (methacetin breath test) and Portal Hepatic Filtration rate (HepQuant)

Results

Safety Summary

- SZN-043 was well tolerated across all populations and dosing regimens. No clinically significant vital sign changes, ECG nor physical exam findings were noted for any participants. No SUSARs nor SAEs were reported during the study. No participants experienced an infusion reaction. The majority of adverse events assessed to be probably related to SZN-043 (and confirmed to have received SZN-043) were unremarkable and limited to 2 participants in Part 3 (MAD) : 1 participant at 1.0 mg/kg experienced a mild headache (on the day of the second dose); 1 participant at 3.0 mg/kg experienced palpitations and rash at the infusion site (occurring before the second dose). All events resolved with no sequalae. Product administration and/or phlebotomy procedure related events were also reported in 11 different participants with no appreciable difference between placebo and treated participants, nor pattern around dose or frequency of SZN-043 administered.
- The only adverse events of interest were an unexplained rise in serum transaminases that were unanticipated from nonclinical data observed in eight (8) HVs, although two in the multiple dose cohorts (one at each dose) were considered unrelated by the investigator. Measurements of ALT and AST found elevations as high as grade 2 at a rate higher than in placebo participants. Transaminase elevations appeared dependent on magnitude of any single dose rather than cumulative doses. No rises in transaminases were observed in PHLCs. The events are summarized below.

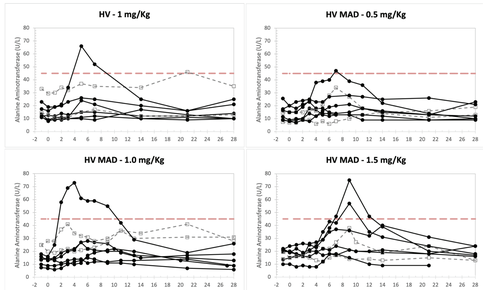
Transaminase elevations (Table 1, Figures 3, 4)

- Transaminase elevations were observed in a subset of SZN-043 treated subjects which:
  - Were transient with incidence that was dose related, with interpatient variability
  - Resolved without intervention
  - Were not associated with other changes in clinical pathology (e.g. GGT)
  - Were confirmed as hepatocyte-related as evidenced by concomitant increases in serum levels of GLDH and CK18

Table 1. Summary of transaminase changes

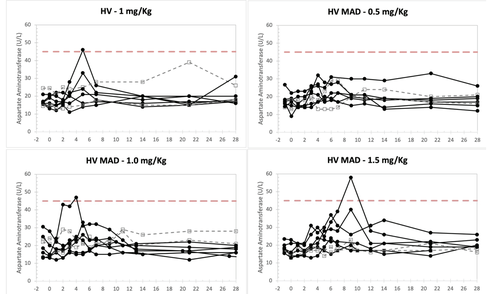
	1 mg/kg	3 mg/kg	0.5 mg/kg	1 mg/kg	0.5 mg/kg	1 mg/kg	1.5 mg/kg
Part of Study	Part 1 (SAD)		Part 2 (SAD)		Part 3 (MAD) Day 0, Day 4		
Population	Healthy Volunteers		Patients with history of cirrhosis		Healthy Volunteers		
SZN-043/ Placebo	6/2	6/2	3/1	3/1	6/2	6/2	6/2
Grade 2+ Transaminase Elevations	0	2 Grade 2	0	0	0	0	0
Grade 1 Transaminase Elevations	2	1	0	0	0	1	2

Figure 3. Serum ALT concentrations



Solid lines = SZN-043 treated subjects; Dotted lines = placebo subjects

Figure 4 Serum AST concentrations

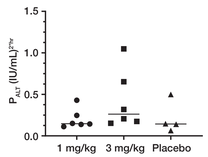


Solid lines = SZN-043 treated subjects; Dotted lines = placebo subjects

P_{ALT}

- P_{ALT} values[1] based on ALT elevations were well below 5, indicating that hepatocyte loss associated with SZN-043 administration may be considered not clinically significant (Figure 5)

Figure 5. P_{ALT} calculations



Investigations to Assess Mechanism of Transaminase Elevations

- No consistent evidence of transaminase increases in nonclinical studies, including GLP toxicology studies in two species at substantially higher doses.
- Unable to demonstrate direct hepatotoxicity in any in vitro or in vivo test system
- Hypotheses explored and excluded:
  - Immune mediated hepatotoxicity
  - Upregulation of transaminase formation by hepatocytes
  - Bile acid dysregulation
  - Heparin mediated hepatotoxicity
  - Fasting induced refeeding
- CYP2E1 induction resulting in oxidative stress (OS) and hepatocellular injury
  - Studies in primary hepatocytes not able to demonstrate OS or cellular injury under multiple experimental conditions
  - Serum isoprostanes, biomarker for OS, unchanged in SZN-043 treated subjects

Pharmacokinetics of SZN-043

- The pharmacokinetics of SZN-043 were consistent with an IgG-based therapeutic binding to a high abundance target (Figure 6, Table 2)
  - Elimination decreases with increasing dose
  - C_{max} proportional to dose
- Repeated dosing does not result in accumulation or changes in PK

Figure 6. Mean (SD) serum SZN-043 concentrations

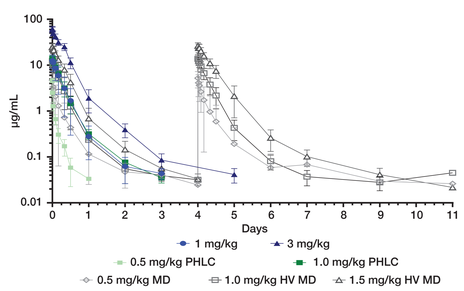


Table 2. Mean (SD) pharmacokinetic parameters of SZN-043

Dose (mg/kg)	Single dose, HV		Multiple dose, HV Days 0, 4			Single dose, PHLC	
	1	3	0.5	1	1.5	0.5	1
AUC (µg-day/mL)	3.2 (1.9)	34.9 (6.6)	2.09 (1.81)	6.73 (2.31)	17.6 (4.67)	0.475 (0.145)	3.34 (0.893)
CL (mL/day/kg)	454 (324)	89.0 (19.8)	734 (450)	327 (112)	181 (48.1)	1110 (310)	312 (72.3)
Terminal half-life (Days)	0.737 (0.218)	3.40 (1.27)	1.06 (1.09)	1.29 (1.34)	1.37 (0.285)	0.346 (0.160)	0.638 (0.109)
C_{max} (µg/mL)	12.6 (4.12)	61.9 (8.25)	4.68 (1.74)	12.9 (1.63)	26.4 (4.47)	4.61 (0.477)	14.1 (1.90)
V_c (mL/kg)	85.0 (27.2)	48.7 (6.81)	113 (50.3)	85.6 (10.3)	61.8 (11.4)	96.4 (9.12)	69.1 (9.91)

AUC = area under the concentration vs time curve from 0-infinity; CL = clearance; C_{max} = maximum observed serum concentration; Vc = central compartment volume of distribution

Antidrug Antibodies (ADA)

- 12 of 36 (33%) of SZN-043 treated subjects showed treatment-emergent anti-drug antibodies against SZN-043. Of these, only one subject exhibited a transiently positive ADA response against RSPO2.
- The was no discernable effect of ADA on SZN-043 exposure; nor was there any pharmacologic evidence of neutralization of endogenous RSPO2.

Pharmacodynamic biomarkers

- Binding to, and elimination of, ASGPR from the surface of hepatocytes by SZN-043 was demonstrated by an increase in serum ALP (Figure 7). This transient, benign elevation occurs as a result of reduced clearance of ALP by ASGPR.
- An increase in methacetin clearance following SZN-043 administration is indicative of Wnt-mediated upregulation of CYP1A2 in hepatocytes (Figure 8)
- Portal hepatic filtration (HFR) as measured by increased clearance of d4-cholate using HepQuant, is thought to be related to Wnt-mediated induction by SZN-043 of hepatic uptake transporters (Figure 9)2

Figure 7. Mean (SD) percent change from baseline in serum ALP

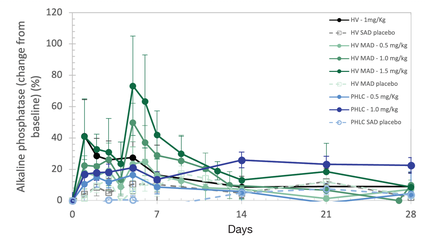
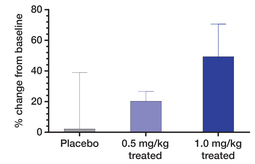
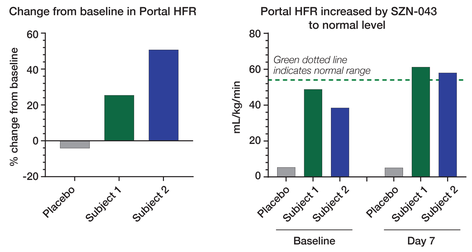


Figure 8. CYP1A2 mediated clearance of methacetin (Mean/SD) in PHLC subjects



Percent change from baseline to 3 days after SZN-043 administration.

Figure 9. Portal HFR (PHLC dosed at 1 mg/kg)



Summary

- Wnt modulation in hepatocytes is a promising new mechanism for supporting regeneration in injured livers
- SZN-043 is a novel biotherapeutic shown to potentiate Wnt signaling and induce proliferation in hepatocytes in mice
- In a Phase Ia study in HVs and PHLCs, single and multiple doses of SZN-043 were safe and well tolerated
- Mild-to-moderate, transient, dose-related serum transaminase elevations were noted in some treated subjects
- Serum SZN-043 exposure was consistent with an IgG-based fusion protein.
- Pharmacodynamic responses indicating target occupancy and hepatic Wnt-mediated signaling were observed
- The results from this study warrant further clinical investigation of SZN-043. A Phase Ib study in subjects with severe alcohol-associated hepatitis is actively recruiting.

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