Safety, Tolerability, and Pharmacokinetics of GS-9450 in Healthy Male and Female Volunteers

44th Annual Meeting of the
European Association for the Study of the Liver
April 22 - 26, 2009
Copenhagen, Denmark

F. Höppener[1], J.A. Kim[2], M.J. Park[2], and H.J. Choi[2]

[1]KENDLE Clinical Pharmacology Unit, Utrecht, The Netherlands; [2]LG Life Sciences, Ltd., Seoul, Republic of Korea

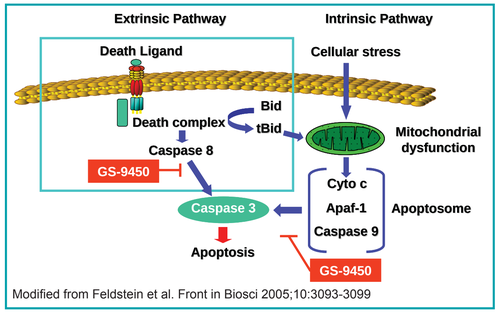
LG Life Sciences

LG Life Sciences, Ltd.
20, Yoido-dong, Youngdungpo- gu,
Seoul, 150-721, Republic of Korea
Tel: 82-2-3773-3966
Fax: 82-2-785-0324

Introduction

- Apoptosis, or programmed cell death, is characterized by distinctive morphological changes such as cell shrinkage and nuclear condensation
- Biochemically, a hallmark of apoptosis is the activation of caspases, which are a structurally related family of cysteine aspartyl proteases responsible for cleavage of several critical cellular targets
- Caspases can be controlled upstream by the regulation of signals that lead to zymogen activation, or downstream by inhibitors that prevent them from reaching their substrates
- Activation of caspase 8 by apoptotic signaling pathways including Fas and TNF-α, in turn, activates other apical caspases (e.g., caspase 9) or "executioner" caspases (e.g., caspases 3, 6, and 7) that ultimately target a number of cellular components.
- This caspase cascade results in the transformation of a functioning cell into an apoptotic body. Although low levels of apoptosis normally occur to maintain homeostasis, abnormal amounts of apoptosis occur in disease states, especially in the liver

Figure 1. Apoptosis Pathways in Liver Disease



Modified from Feldstein et al. Front in Biosci 2005;10:3093-3099

Background

- Activation of caspases and eventual cell apoptosis has been associated with a number of liver diseases, including non-alcoholic and alcoholic steatohepatitis, chronic hepatitis B/C virus infection, and cholestatic liver injury
- This suggests that caspase inhibitors may be therapeutically useful in these diseases. GS-9450 was found to be a very potent inhibitor of caspase activity during in vitro and in vivo pre-clinical studies. These first-in human clinical studies have been performed to assess safety, tolerability and pharmacokinetics (PK) of GS-9450 in healthy volunteers before exploring its therapeutic value in patient groups

Objectives

- To determine the safety, tolerability and pharmacokinetics of GS-9450 following administration of single and multiple, escalating oral doses of GS-9450 in healthy male subjects and to assess the safety, tolerability and the pharmacokinetics of GS-9450 following administration of a single oral dose in a single cohort of female subjects
- To preliminarily assess the effect of food on the pharmacokinetics of GS-9450 in healthy male subjects

Methods

- Two, randomized, double-blind, placebo-controlled Phase I studies were performed exposing 54 healthy male subjects and 6 healthy female subjects to GS-9450
- Healthy male subjects received GS-9450 doses of 5 to 120 mg in a single ascending dose (SAD) study and 40 to 120 mg/day for 2 weeks in a multiple ascending dose (MAD) study; female subjects received a single GS-9450 dose (SD) of 40 mg
- To evaluate the effect of food intake on the pharmacokinetics of GS-9450, subjects participating in the 40 mg SAD cohort received a second dose of GS-9450 after ingestion of a high fat breakfast
- Blood and urine samples were analyzed to determine GS-9450 and metabolite concentrations in order to calculate single dose and steady state PK parameters
- Safety and tolerability were assessed by means of recording of adverse events (AEs) vital signs measurements, 12-lead ECG recordings, clinical laboratory tests and physical examinations

Results

Demographics Summary

[SAD] Age range: 18 ~ 55 years
Race distribution: 1 Asian, 2 Afro-Europeans, 45 Caucasians
[MAD] Age range: 20 ~ 53 years (MAD), 20 ~ 51 years (Female SD)
Race distribution: 1 Hispanic female, 2 Afro-Europeans, 29 Caucasians

Table 1. Demographic Characteristics (SAD)

Dose of GS-9450		0 mg (N = 12)	5 mg (N = 6)	10 mg (N = 6)	20 mg (N = 6)	40 mg (N = 6)	80 mg (N = 6)	120 mg (N = 6)	40 mg/fed (N = 4)
Age	Mean	29.8	32.0	25.2	25.2	28.7	42.2	40.5	28.8
(years)	SD	12.8	17.8	6.11	9.11	11.9	13.6	15.6	13.3
Height	Mean	181	181.8	186	178.7	184.2	176.8	178.8	182.1
(cm)	SD	8.13	9.256	3.27	6.322	8.699	6.548	5.871	9.724
Weight	Mean	76.67	79.47	80.82	71.92	76.33	75.1	74.3	72.45
(kg)	SD	10.61	10.58	8.503	9.051	15.07	7.09	8.68	8.052
BMI	Mean	23.4	24.0	23.3	22.5	22.5	24	23.0	22.0
(kg/m^2)	SD	3.06	2.37	2.25	2.88	3.15	2.9	2.28	1.41

Table 2. Demographic Characteristics (Female SD)

Dose of GS-9450		Female / SD	
		0 mg (N = 2)	40 mg (N = 6)
Age	Mean	23.5	27.8
(years)	SD	4.95	11.6
Height	Mean	167.5	167.9
(cm)	SD	4.950	7.039
Weight	Mean	63.60	64.07
(kg)	SD	1.980	6.706
BMI	Mean	22.72	22.8
(kg/m^2)	SD	2.048	3.27

Table 3. Demographic Characteristics (MAD)

Dose of GS-9450		MAD			
		0 mg (N = 6)	40 mg (N = 6)	80 mg (N = 6)	120 mg (N = 6)
Age	Mean	40.8	33.5	39.7	32.5
(years)	SD	9.50	9.89	10.2	10.9
Height	Mean	178.7	181.2	183.5	178.3
(cm)	SD	7.960	4.155	3.742	4.107
Weight	Mean	79.87	81.57	83.70	84.88
(kg)	SD	9.952	10.27	6.436	6.971
BMI	Mean	24.95	24.97	24.87	26.70
(kg/m^2)	SD	1.664	3.924	1.893	2.105

Safety Summary

- Overall, GS-9450 was well tolerated with no serious (SAEs) or severe adverse events reported
- The most frequently reported AE was headache
- All treatment-emergent AEs were transient, and most AEs were mild
- There was no evidence for a dose-related increase in the incidence or severity of AEs
- One male subject discontinued dosing during the MAD study due to development of a rash of mild severity.
- No effects were observed on vital signs measurements, ECG recordings, continuous cardiac monitoring or safety laboratory test results

Results *(cont'd)*

Table 4. Summary of Possibly or Probably Drug-related AEs (SAD)

GS-9450 Treatment System Organ Class Preferred Term	0 mg (N = 12) n (%)	5 mg (N = 6) n (%)	10 mg (N = 6) n (%)	20 mg (N = 6) n (%)	40 mg (N = 6) n (%)	80 mg (N = 6) n (%)	120 mg (N = 6) n (%)	40 mg/ (N = 4) n (%)
Gastro-intestinal	-	-	-	1 (16.7)	1 (16.7)	-	-	1 (25.0)
Abdominal pain	-	-	-	1 (16.7)	1 (16.7)	-	-	-
Dry throat	-	-	-	-	-	-	-	1 (25.0)
Musculo skeletal/ connective tissue	1 (8.3)	-	-	-	1 (16.7)	-	-	-
Myalgia	1 (8.3)	-	-	-	1 (16.7)	-	-	-
Nervous system	2 (16.7)	1 (16.7)	1 (16.7)	1 (16.7)	-	-	-	-
Dizziness	-	-	1 (16.7)	-	-	-	-	-
Headache	2 (16.7)	1 (16.7)	1 (16.7)	-	-	-	-	-
Somnolence	-	-	-	1 (16.7)	-	-	-	-
Renal and urinary	-	-	-	-	-	1 (16.7)	-	-
Micturition	-	-	-	-	-	1 (16.7)	-	-
TOTAL	2 (16.7)	1 (16.7)	1 (16.7)	2 (33.3)	2 (33.3)	1 (16.7)	-	1 (25.0)

Table 5. Summary of Possibly or Probably Drug-related AEs (MAD / Female SD)

GS-9450 Treatment System Organ Class Preferred Term	MAD				Female SD	
	0 mg (N = 6) n (%)	40 mg (N = 6) n (%)	80 mg (N = 6) n (%)	120 mg (N = 6) n (%)	0 mg (N = 2) n (%)	40 mg (N = 6) n (%)
Gastrointestinal	2 (33.3)	1 (16.7)	1 (16.7)	1 (16.7)	-	-
Abdominal pain	2 (33.3)	1 (16.7)	-	-	-	-
Constipation	-	-	1 (16.7)	-	-	-
Dry throat	-	-	1 (16.7)	-	-	-
Nausea	-	-	-	1 (16.7)	-	-
General/ administration site conditions	-	-	1 (16.7)	1 (16.7)	-	-
Fatigue	-	-	1 (16.7)	-	-	-
Malaise	-	-	-	1 (16.7)	-	-
Musculoskeletal and connective tissue	-	-	2 (33.3)	-	-	-
Back pain	-	-	1 (16.7)	-	-	-
Neck pain	-	-	1 (16.7)	-	-	-
Nervous system	5 (83.3)	1 (16.7)	1 (16.7)	2 (33.3)	1 (50.0)	1 (16.7)
Headache	2 (50.0)	1 (16.7)	1 (16.7)	2 (33.3)	1 (50.0)	1 (16.7)
Pharyngeal hypoaesthesia	1 (16.7)	-	-	-	-	-
Somnolence	2 (33.3)	-	-	-	-	-
Skin and subcutaneous tissue	1 (16.7)	1 (16.7)	-	-	-	-
Pruritus generalized	1 (16.7)	-	-	-	-	-
Rash	-	1 (16.7)	-	-	-	-
TOTAL	6 (100.0)	2 (33.3)	3 (50.0)	3 (50.0)	1 (50.0)	1 (16.7)

N = number of subjects in specified group, n = number of subjects with AEs, % = n/N x 100%

Pharmacokinetics Summary

- C_{max} was attained at 0.5 to 3 hours (hrs) post-dose
- Mean elimination half-lives ($t_{1/2}$) ranged from 11 to 18 hr across all dose levels with a female cohort showing approximately 4 hours longer than those of male cohorts
- Systemic exposure to GS-9450 generally increased in a dose-proportional manner over the range of single and multiple doses
- Only 2~4% of the administered dose was recovered unchanged in the urine
- Neither food nor gender had a significant effect on the PK of GS-9450
- At steady state, there was no significant accumulation of GS-9450 in plasma. The mean accumulation ratio ranged from 1.1 to 1.4 across dose cohorts
- Median t_{max} occurred at 1 to 4 hrs post-dose for the metabolites (GS-9472, GS-9473, GS-9471 and GS-9470) across the dose cohorts at steady state with median $t_{1/2}$ values of metabolites ranging from 13 to 18 hrs
- Overall, the arithmetic mean (C_{max} and AUC_{tau}) of the metabolites were proportional to the dose over the dose range of 40 to 120 mg, suggesting metabolism of GS-9450 is independent of dose

Figure 2. Plasma Concentration Time Profile of GS-9450 (SAD)

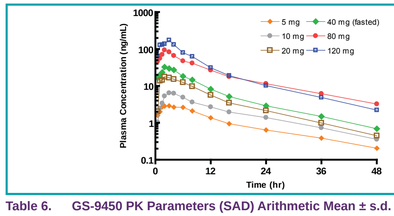


Table 6. GS-9450 PK Parameters (SAD) Arithmetic Mean ± s.d.

Dose (mg)	N	Condition	Sex	t_{max}[a] (hr)	C_{max} (ng/mL)	AUC_{0-t} (hr*ng/mL)	$t_{1/2Z}$ (hr)	CL/F (L/hr)
5	6	fasted	M	2.5 (1.5~6.0)	3.5±0.7	51.40±14.09	15.1±5.2	104±31
10	6	fasted	M	3.0 (2.0~4.0)	8.2±1.3	95.98±20.70	12.0±1.7	108±20
20	6	fasted	M	2.25 (1.0~4.0)	21.4±8.1	204.7±61.29	10.9±2.0	105±32
40	6	fasted	M	2.0 (0.5~4.0)	44.7±10.1	324.5±76.03	11.4±1.2	129±28
40	4	fed	M	3.5 (3.0~6.0)	43.4±15.2	420.8±79.71	13.0±1.9	98±18
40	6	fasted	F	2.50 (1.5~4.0)	45.7±12.7	374.3±100.7	17.5±2.6	114±33
80	6	fasted	M	2.0 (1.0~3.0)	109.8±29.2	1032±319.4	12.2±3.8	83±23
120	6	fasted	M	3.0 (1.0~3.0)	203.5±117.5	1401±488.0	10.7±1.9	96±37

a. Median value (min~max)

Figure 3. Plasma Concentration-Time Profile of GS-9450 (MAD, Day 1 vs. Day 16)

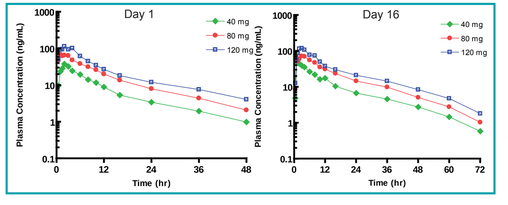


Table 7. GS-9450 PK Parameters (MAD)

Dose (mg)	N	Regimen	t_{max}[a] (hr)	C_{max} (ng/mL)	AUC_{0-t} (hr*ng/mL)	$t_{1/2Z}$ (hr)	CL/F (L/hr)
40	6	Day 1	2.0 (1.0~4.0)	37.87±3.48	335.1±48.9	13.4±3.6	115.0±18.1
		Day 16	0.5 (0.5~1.0)	54.88±8.89	371.9±41.0	15.6±3.2	108.6±12.1
80	6	Day 1	1.5 (0.5~4.0)	90.50±52.16	740.5±259.2	12.0±1.2	113.1±36.1
		Day 16	2.0 (0.5~6.0)	86.62±16.02	714.0±168.8	15.8±2.3	107.1±36.3
120	6	Day 1	2.5 (1.5~4.0)	146.8±34.8	1139±169.8	15.3±1.9	99.5±14.4
		Day 16	1.75 (0.5~6.0)	138.8±39.2	1028±190.3	16.5±2.6	119.8±20.4

a. Median value (min~max)

Figure 4. Plasma Concentration-Time Profile of GS-9450 40 mg SD Male vs. Female

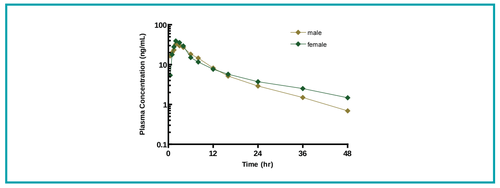
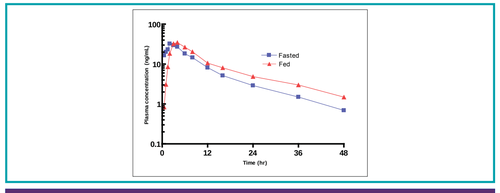


Figure 5. Plasma Concentration-Time Profile of GS-9450 40 mg SD Fasted vs. Fed



Conclusions

- **Single oral doses of 5 to 120 mg and multiple oral doses of 40 to 120 mg of GS-9450 were well tolerated by healthy male subjects and single oral doses of 40 mg were well tolerated by female subjects**
- **The half-life GS-9450 supports once-daily dosing and no evidence was found for a significant gender or food effect on GS-9450 PK**
- **In conclusion, the results from these Phase I studies combined with the *in vivo* efficacy data make GS-9450 a promising candidate for further development as treatment for hepatic diseases, in which the pathogenesis is mediated through apoptosis. Phase II trials with GS-9450 are currently being performed in chronic HCV infected and NASH patient populations**

Acknowledgements

The authors would like to thank the volunteer participants and the staff at Kendle Clinical Pharmacology Unit, Utrecht, Netherlands for their participation in this study.

For additional information on GS-9450 please also see poster #969