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PRECLINICAL PHARMACOKINETICS OF CK-4021586, A NOVEL INHIBITOR OF CARDIAC MYOSIN, AND PREDICTION OF HUMAN PHARMACOKINETICS

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PURPOSE

CK-4021586 is a novel small molecule cardiac myosin inhibitor in early-stage clinical development that was designed as an orally administered drug to treat pathological hypercontractility leading to hypertrophy, fibrosis, and left ventricular diastolic dysfunction in patients with heart failure with preserved ejection fraction (HFpEF).

The purpose of the present studies was the characterization of **CK-4021586** preclinical pharmacokinetics toward the prediction of human pharmacokinetics using a range of methods.

OBJECTIVE

The characterization of **CK-4021586** preclinical pharmacokinetics toward the prediction of human pharmacokinetics using a range of methods.

METHODS

The pharmacokinetics of **CK-4021586** was evaluated in single intravenous dose studies in the mouse, rat, dog and monkey. The predicted $t_{1/2}$ of **CK-4021586** in human was calculated using the average of predicted human pharmacokinetic CL and Vss parameters using a range of methods described below and the relationship, $t_{1/2} = V_{ss}*0.693/CL$. **Human oral bioavailability and absorption rate constant predictions** were determined from pharmacokinetic studies using pentagastrin-treated beagle dogs dosed orally with 20 mg/kg of non-micronized **CK-4021586** in a gelatin capsule.

Human clearance (CL) was estimated from an average of seven different CL prediction methods:

1. 4-Species simple allometric scaling of total CL with maximum life span potential rule of exponents correction;
2. Liver blood flow-based scaling from rat, dog, and monkey;
3. Two species rat-dog multi-linear regression;
4. Two species rat-dog allometry;
5. Two species rat-monkey allometry;
6. Fraction unbound corrected intercept method (FCIM)
7. Human liver microsomal CL *in vitro*-to-*in vivo* extrapolation (IVIVE) using the well-stirred model with corrections for both plasma protein binding and incubation non-specific binding

Human volume of distribution (V_{ss}) was estimated from an average of five different methods:

1. 4-Species simple allometric scaling of unbound V_{ss};
2. Two species rat-dog allometry;
3. Monkey as a single species (unbound);
4. Øie-Tozer method using all 4-species;
5. Arundel model using rat, dog, and monkey

RESULTS

Figure 1. Cross-species Concentration-time Profiles of CK-4021586 Post-administration Intravenous or Oral. Values are Mean ± SD, n=3.

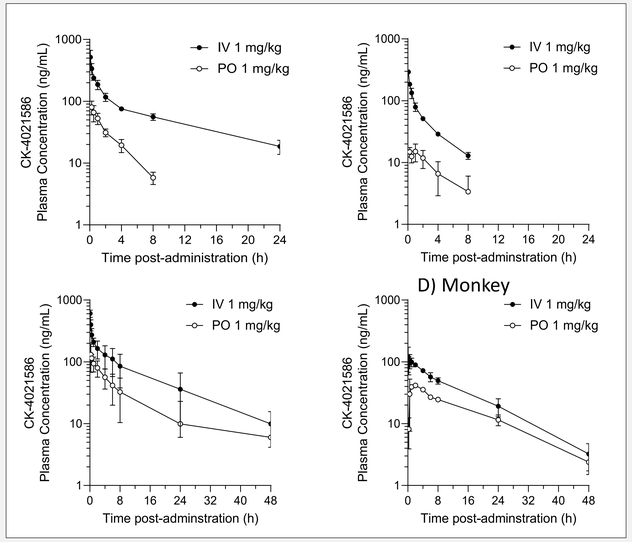

Table 1. CK-4021586 Pharmacokinetic Summary in Pre-clinical Species Following Intravenous or Oral Administration.

Preclinical species CK-4021586 intravenous administration pharmacokinetics (Male, Fed, Mean [N=3 animals])				
Species/Strain	**Dose[a] (mg/kg)**	**CL (mL/min/kg)**	**V_{ss} (L/kg)**	**$t_{½}$ (h)**
Mouse/C57BL/6	1	9.33	6.54	10.2
Rat/Sprague Dawley	1	37.3	7.67	3.11
Dog/beagle	1	7.57	4.23	9.54
Monkey/cynomolgus	1	11.6	8.89	9.94

CL = plasma clearance; $t_{½}$ = terminal half-life; V_{ss} = apparent volume of distribution at steady-state
[a]The vehicle used was 10% DMA/40% PG/50% HPβCD (40% w/v aqueous solution).

Preclinical species CK-4021586 oral administration pharmacokinetics (Male, Fasted, Mean [N=3 animals])				
Species/Strain	**Dose[a] (mg/kg)**	**t_{max} (h)**	**F (%)**	**$t_{½}$ (h)**
Mouse/C57BL/6	1	0.25	12.6	2.47
Rat/Sprague Dawley	1	0.50	16.5	3.30
Dog/beagle	1	0.33	35.2	7.46
Monkey/cynomolgus	1	1.17	51.4	11.7

F = oral bioavailability; M = male; t_{max} = time to maximum plasma concentration
[a]Oral gavage administration. The vehicle used was 0.5% HPMC/0.1% Tween 80 in water

Figure 2. CK-4021586 Simple Allometric Scaling of Interspecies A) Total Plasma CL (with maximum lifespan, MLP, correction), and B) Unbound Plasma V_{ss} Normalized to Body Weight.

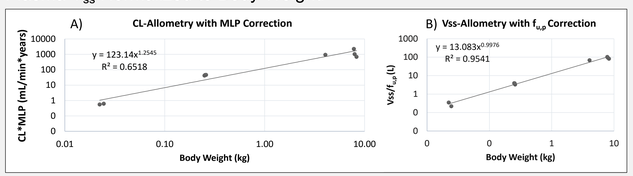

Figure 3. Disappearance of CK-4021586 Following Incubations of 1 µM CK-4021586 with 1 mg/mL Rat (RLM), Dog (DLM), Cynomolgus Monkey (CLM), and Human (HLM) Liver Microsomes.

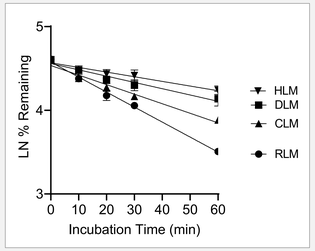

Table 2. Metabolic Stability of CK-4021586 in Rat, Dog, Monkey, and Human Liver Microsomes and Prediction of *In Vivo* Hepatic Clearance.

Parameter	Rat	Dog	Monkey	Human
Incubation slope (k)	-0.0179	-0.0075	-0.0114	-0.0056
Incubation half-life (min)	38.7	92.2	60.7	124
Microsome factor (mg/g liver)	45	45	45	45
(Liver g/body kg)	40	32	30	25.7
Fraction unbound in plasma ($f_{u,p}$)	0.529	0.361	0.577	0.415
Fraction unbound in incubation ($f_{u,inc}$)	0.320	0.340	0.320	0.330
$f_{u,p}/f_{u,inc}$	1.7	1.1	1.8	1.3
Hepatic blood flow (Q_h)	55.2	30.9	43.6	20.7
CL_{int} (mL/min/mg liver microsomal protein)	0.0179	0.0075	0.0114	0.0056
CL_{int} (mL/min/kg)	32.3	10.8	15.4	6.4
Predicted $CL_{H,blood}$ (mL/min/kg)	27.1	8.4	17.0	5.8
Blood-to-plasma ratio	0.95	0.86	1.03	0.93
*Predicted $CL_{H,plasma}$ (mL/min/kg)	25.7	7.19	17.4	5.43
Observed CL_{plasma} (mL/min/kg)	37.3	7.57	11.6	-
Observed CL_{plasma}/Predicted $CL_{H,plasma}$	1.5	1.1	0.7	-

*Predicted $CL_{H,plasma}$ = Predicted $CL_{H,blood}$ multiplied by the corresponding blood-to-plasma ratio

Figure 4. Mean Plasma Concentration-Time Profile for CK-4021586 in Male Pentagastrin-treated Beagle Dogs Following PO Dosing at 20 mg/kg Using Capsule Formulation (Mean ± SD, N=3) (the inset shows simulated pharmacokinetic profile for the estimation of K_a (K01).

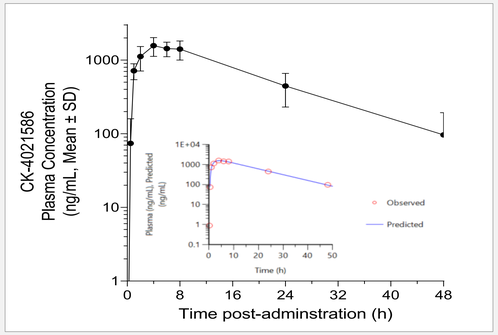

Table 3. Methods Used in the Calculation of Predicted CK-4021586 Pharmacokinetic Parameters in Human.

Plasma Clearance (mL/min/kg)		**RESULTS**
Simple allometry (MLP)	$CL*MLP = a(BW)^b$	3.9
LBF (rat, dog, monkey)	$CL/f_{u,p}(human) = CL/f_u(animal)*(human/animal)Q_h$	6.1
Rat-dog MLR	$log(CL_{human}) = 0.4*log(CL_{iv,rat}) + 0.4*log(CL_{iv,dog}) - 0.4$	3.8
Rat-dog allometry	$A_{rat\text{-}dog}(BW_{human})^{0.628}$	4.1
Rat-monkey allometry	$A_{rat\text{-}monkey}(BW_{human})^{0.650}$	4.7
FCIM	$CL = 33.35*(a/Rf_{u,p})^{0.77}$	2.9
IVIVE Well-stirred model	$CL_h = ((f_{u,p}*CL_{int}/f_{u,inc})*Q_h)/((f_{u,p}*CL_{int}/f_{u,inc}) + Q_h)$	5.4
Average		**4.3**
Plasma V_{ss} (L/kg)		**RESULTS**
Simple allometry, unbound	$V_{ss}/f_{u,p} = a(BW)^b$	5.4
Rat-dog allometry	$log(V_{human}) = (0.07714*logV_{rat}*logV_{dog}) + (0.5147*logV_{dog}) + 0.586$	3.5
Monkey single-species (unbound)	$V_{human} = V_{monkey}$ & $V_{human}/f_{u,p} = V_{monkey}/f_{u,p}$	6.4
Arundel model (rat, dog, monkey)	$V_{ss} = V_p + \Sigma (V_t*K_{put}*f_{u,p})$	5.0
Øie-Tozer (4-species)	$V_{ss} = V_p + (f_{u,p,human}*V_e) + [(1\text{-}f_{u,p,human})*(R_{e/i})*V_p] + V_r*(f_{u,p,human}/f_{u,t,human})$	8.0
Average		**5.8**
Human Half-life (h)	$t_{1/2} = [ln2]*V_{ss}/CL$ (assuming 1-compartment pharmacokinetics)	**15.6**
Human Bioavailability (pentagastrin dog non-micronized drug in capsule, 200 mg): **%F**		**50**
Human Absorption rate constant (pentagastrin dog, drug in capsule, 200 mg): **K_a, 1/h**		**0.68**

CONCLUSIONS

- A combination of allometric scaling and IVIVE methods, which employed the use of *in vivo* preclinical pharmacokinetics and *in vitro* human liver microsome CL evaluation, were utilized for the prediction of **CK-4021586** human pharmacokinetic parameters.
- The **CK-4021586** mean predicted human plasma CL and V_{ss} are **4.3 mL/min/kg** and **5.8 L/kg**, respectively, together yielding an estimated human half-life of approximately **15.6 hours**.
- The estimated **CK-4021586** human oral bioavailability was **50%** with an absorption rate constant of **0.68 1/h** from oral PK testing in pentagastrin-treated beagle dogs.

DISCLOSURES AND ACKNOWLEDGMENTS

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