Modeling of Cyclosporine A-Induced Acute Kidney Injury with RENAsym®

Jeffrey L. Woodhead[a], Pallavi Bhargava[a], Christina Battista[a], Viera Lukacova[b]

[a]DILIsym Services Division, Simulations Plus Inc., Research Triangle Park, North Carolina

[b]Simulations Plus Inc., Lancaster, California

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ABSTRACT

Background: Cyclosporine A (CsA) is an immunosuppressant commonly used to prevent organ rejection and can be used to treat other diseases such as rheumatoid arthritis, atopic dermatitis, and psoriasis[1]. However, the use of CsA can cause tubular damage leading to a decline in renal function as indicated by increases in serum creatinine levels and decreases in glomerular filtration rate (GFR)[2]. Low (2-8 mg/kg) and high (10-17 mg/kg) doses have the potential to cause lasting renal injury[1]. This work uses RENAsym, a quantitative systems toxicology (QST) model of acute kidney injury (AKI), to recapitulate clinical outcomes following low and high dose CsA administration in humans.

Methods: To define to the potential for CsA-induced nephrotoxicity, the effects of CsA on mitochondrial function and reactive oxygen species (ROS) production were determined. Human renal proximal tubule epithelial cells (RPTECs) were treated with CsA and its effects on mitochondrial respiration as well as ROS production were measured. The Seahorse XFe96 Analyzer (Agilant, Santa Clara, CA) was used to measure mitochondrial respiration and high content screening was used to measure ROS production after RPTECs were exposed to dihydroethidium staining.

These *in vitro* data were used to determine kidney toxicity parameters for use in RENAsym. CsA inhibited the mitochondrial election transport chain flux and induced ROS production. Together with these kidney toxicity parameters and with physiologically-based pharmacokinetic (PBPK) simulations of low and high clinical CsA exposures, created in GastroPlus®, kidney injury was predicted in RENAsym.

Results: RENAsym predicted CsA-induced kidney injury such as a decrease in kidney average ATP when simulated with 4 mg/kg administration for 24 hours. CsA administration, 4 mg/kg, was simulated for 6 months in RENAsym where an average of 1.5 to 3% decrease in GFR and a 1.5 to 2.5% increase in serum creatinine was predicted in a simulated population (SimPops™) consisting of N=267 individuals. Moreover, a 17 mg/kg QD treatment for 3 months followed by 10 mg/kg QD for another 3 months, was also simulated in RENAsym for 6 months where an average of 4 to 7% decrease in GFR and a 4 to 6% increase in serum creatinine was predicted in the same SimPops.

Conclusions: These data show qualitative agreement with clinical studies of CsA administration showing a decline in renal function[3,4], however further refinement of this representation is underway for RENAsym v2A. This work shows the potential for RENAsym to accurately predict clinical outcomes and nephrotoxicity for future compounds.

INTRODUCTION

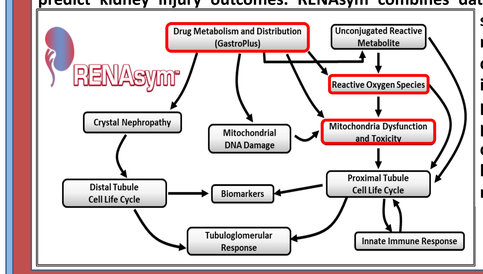
- CsA is an immunosuppressant known for inhibiting T-lymphocyte driven immune responses
- CsA is commonly used following organ transplant to prevent organ rejection and in other diseases such rheumatoid arthritis, atopic dermatitis, and psoriasis
- Treatments of CsA, both low (2-8 mg/kg) and high doses (10-17 mg/kg), can cause renal tubular damage subsequently leading to a decrease in renal function[2]
- Long-term treatment of 4 mg/kg BID for several years caused decreases in GFR and irreversible kidney damage based on renal histology[3]
- A high-dose treatment of 17 mg/kg QD for 3 months followed by 10 mg/kg QD for another 3 months were reported in the literature to increase serum creatinine levels > 2mg/dL and decrease GFR by at least 40%[4,5]
- Here we simulate both low and high dose treatments of CsA and qualitatively recapitulate clinical outcomes observed in the literature

DILI-sim Initiative

RESULTS

RENAsym Overview

Figure 1: RENAsym is comprised of submodels that interact with one another to predict kidney injury outcomes. RENAsym combines data from *in vitro* toxicity studies, predictions of metabolism and distribution, as well as inner workings of kidney physiology to predict the potential for a given drug to induce acute kidney injury or cause nephrotoxicity.

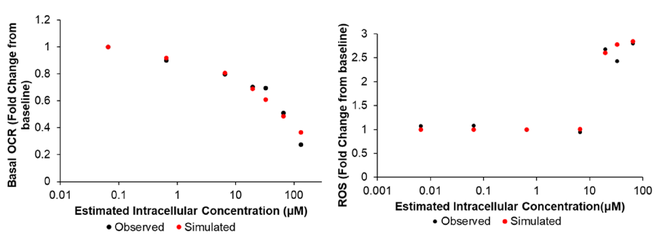


Simulation Scheme

To model CsA-induced nephrotoxicity, an example of a low dose treatment, 4 mg/kg BID for 6 months[3], and high dose treatment, 17 mg/kg QD for 3 months followed by 10 mg/kg for 3 months[4] were simulated. A PBPK model for CsA was available in GastroPlus where both treatments were simulated to predict kidney exposure. The kidney exposure profiles and the toxicity parameters (ROS production and ETC inhibition) for CsA were imported into RENAsym. To determine the effects of CsA on GFR and serum creatinine concentration, both dosing protocols were evaluated in SimPops (N=267). SimPops represent inter-individual variability in a healthy population and variability in susceptibility to toxicity mechanisms such as ROS.

In Vitro Toxicity Parameterization

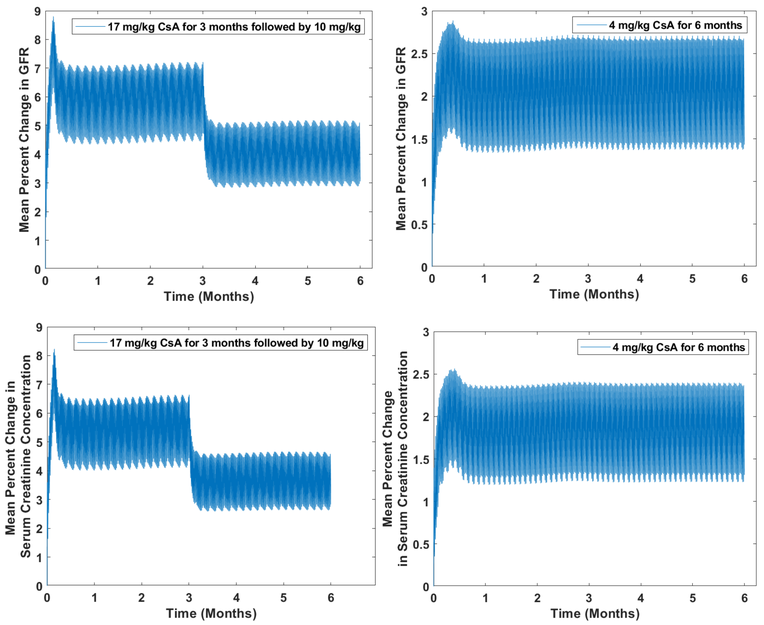
Figure 2: CsA-induced ETC inhibition was simulated in MITOsym (left), and ROS production in RENAsym (middle), against observed mitochondrial respiration and ROS production from *in vitro* studies. Resulting RENAsym toxicity parameter values for CsA are shown in the table (right).



Toxicity Parameter	Value
Non-saturable Coefficient for ETC Inhibition	1458.33 μM
Saturable K_m for ETC Inhibition	1.0488 μM
Saturable V_{max} for ETC Inhibition	0.3878 1/hr
ROS Production V_{max}	0.049 1/hr
ROS Production K_m	13.075 μM

SimPops Simulation Results

Figure 4: Effects of high-dose (left) and low-dose (right) CsA treatment on GFR (top) and serum creatinine concentration (bottom).



GFR and serum creatinine concentration were evaluated in a normal healthy population (N=267). The average percent change for both GFR and serum creatinine concentration is approximately 1-3% for the low-dose treatment and is 4-7% for the high-dose treatment. These results qualitatively capture the decrease in GFR and increase in serum creatinine concentrations according to the literature[3,4].

METHODS

- Human RPTECs were treated with doses of CsA ranging from 0.01 to 25 μM.
- Mitochondrial respiration was measured using a Seahorse XFe96 Analyzer.
- ROS production was measured using high content screening to quantify dihydroethidium staining following CsA exposure.
- PBPK simulations of a low dose (4 mg/kg BID for six months) and high dose (17 mg/kg QD for 3 months followed by 10 mg/kg QD for 3 months) treatments of CsA was simulated in GastroPlus.
- The kidney partition coefficient, Kp, for CsA was used to estimate intracellular concentration in toxicity assays, and toxicity parameterizations were based on estimated intracellular kidney concentration.
- MITOsym® was used to parameterize electron transport chain (ETC) inhibition to *in vitro* mitochondrial respiration studies of CsA. ROS parameterization was performed in RENAsym.
- Simulations predicting kidney function for CsA-induced nephrotoxicity were performed using RENAsym in normal healthy individuals (N=267).

CONCLUSION

- Low and high-dose treatment of CsA was simulated in RENAsym SimPops.
- Our results show qualitative agreement with studies that have reported increases in serum creatinine and decreases in GFR[3,4]
- ATP levels decreased in both treatments according to the dose and an increase in urinary αGST was predicted
- GFR decreased 1-3% when low-dose treatment and 4-7% when high-dose CsA treatment was simulated in individuals
- Serum creatinine concentration increased in both CsA dosing protocols

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