

DILIsym® User Training - DILIsym® Parameters From Data: Bile Acid Transporter Inhibition

DILI-sim Team

*DILIsym® and MITOsym® are registered trademarks of The Hamner Institutes for Health Sciences for computer modeling software and for consulting services.

Goal for This Training Session

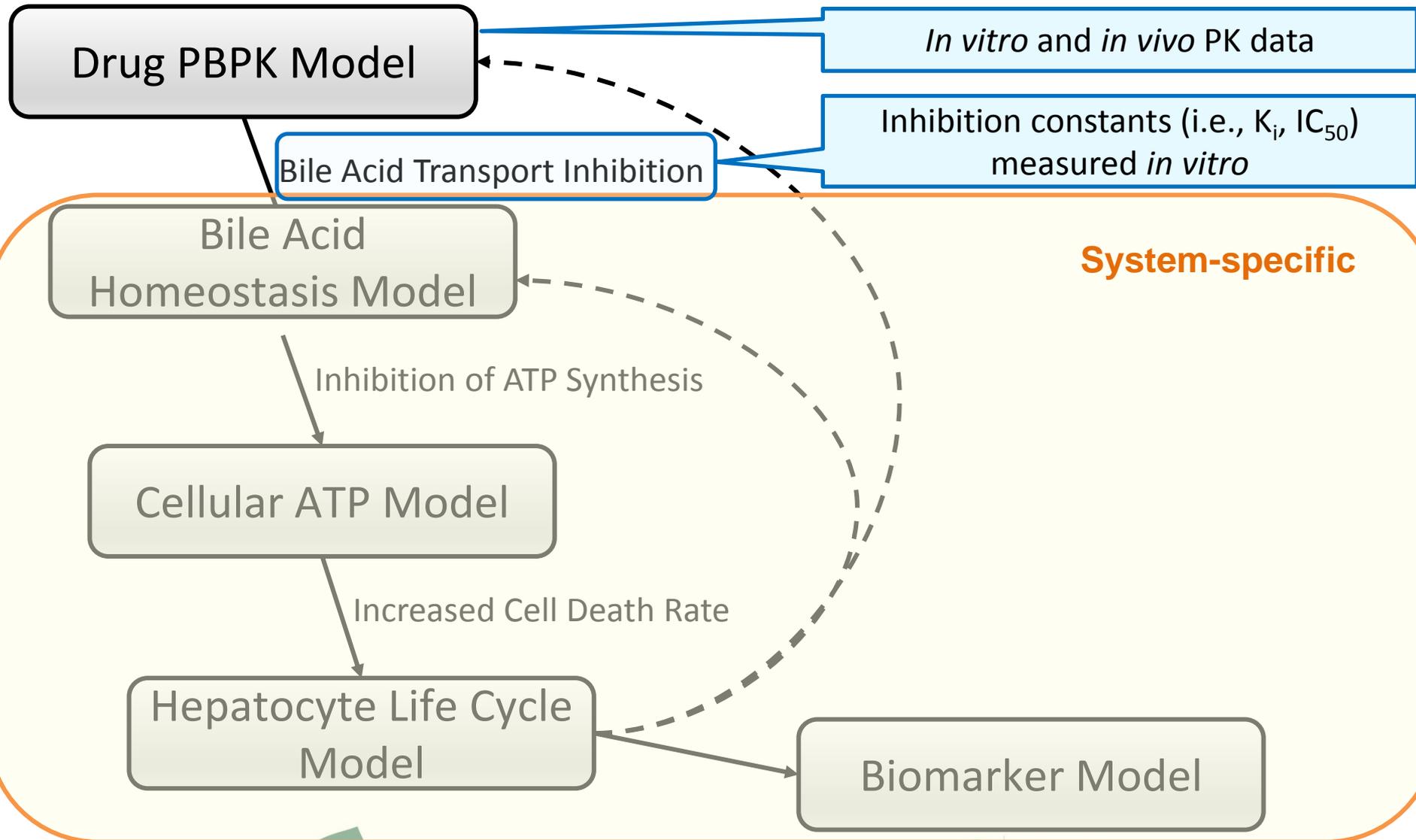
Participants should understand the following general concepts:

- Methods used to parameterize and to simulate bile acid transport disruption in DILIsym[®]

Modeling Compounds that Inhibit Bile Acid Transport: A Case Study with Troglitazone

- Introduction
 - Troglitazone hepatotoxicity was not detected in preclinical studies
 - 2% of patients developed ALT elevations >3X ULN in clinical trials
 - Withdrawn from the market due to idiosyncratic hepatotoxicity
- Modeling troglitazone-mediated hepatotoxicity that involves bile acid transport inhibition
 - Translate data to DILIsym[®] parameters
- Simulate troglitazone-mediated hepatotoxicity using DILIsym[®]
 - Simulate troglitazone-mediated hepatotoxicity in baseline human
 - Simulate troglitazone-mediated hepatotoxicity using human SimPops[™]

Data Inputs for Bile Acid Transport Inhibition



Translate Bile Acid Transport Inhibition Data to DILIsym[®] Parameters for Troglitazone

- Troglitazone competitively inhibits rat Bsep with a K_i of $1.3 \mu\text{M}^\dagger$
 - Will use this value for humans too; literature has shown that troglitazone has similar potency for rat and human BSEP
- Troglitazone inhibits human NTCP and rat Ntcp[‡]
 - IC_{50} values reported: $0.33 \mu\text{M}$ (human), $2.3 \mu\text{M}$ (rat)
 - Type of inhibition not known; assumed to be a competitive inhibitor
- Troglitazone is an inhibitor of human MRP4[§]
 - IC_{50} measured: $21 \mu\text{M}$
 - Type of inhibition not known; assumed to be a non-competitive inhibitor

Troglitazone m.w.
441.5 g/mol

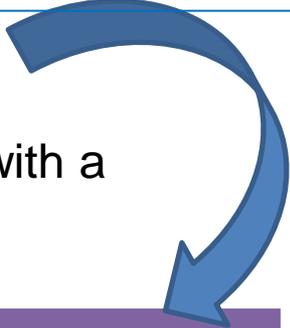
DILIsym [®] Parameter	DILIsym [®] Parameter Input
Compound X BSEP inhibition constant	5.74E-04 mg/mL
Compound X BSEP alpha constant for inhibition	1e10 (competitive)
Compound X BSEP switch	1 (competitive)
Compound X NTCP inhibition constant	1.46E-04 mg/mL (human) 1.02E-03 mg/mL (rat)
Compound X NTCP alpha constant for inhibition	1e10 (competitive)
Compound X NTCP switch	1 (competitive)
Compound X basolateral inhibition constant	9.27E-03 mg/mL
Compound X basolateral alpha constant for inhibition	1 (non-competitive)
Compound X basolateral switch	0 (non-competitive)

[†]Funk 2001, Dawson 2011, [‡]Marion 2007, [§]Yang 2014

Translate Bile Acid Transport Inhibition Data to DILIsym[®] Parameters for Troglitazone Sulfate

- Troglitazone sulfate is a more potent inhibitor of BSEP compared to troglitazone[†]
 - Troglitazone sulfate competitively inhibits rat Bsep with a K_i of 0.23 μM
 - Will use this value for humans too
- Troglitazone sulfate effects on NTCP not known
 - Assumed to be the same as troglitazone[‡]
- Troglitazone sulfate is a non-competitive inhibitor of human MRP4 with a K_i of 8 μM [§]
 - Rat Mrp4 K_i is assumed to be the same as humans

Troglitazone sulfate
m.w. : 521.6 g/mol



DILIsym [®] Parameter	DILIsym [®] Parameter Input
Compound X metabolite B BSEP inhibition constant	1.20E-04 mg/mL
Compound X metabolite B BSEP alpha constant for inhibition	1e10 (competitive)
Compound X metabolite B BSEP switch	1 (competitive)
Compound X metabolite B NTCP inhibition constant	1.46E-04 mg/mL (human) 1.02E-03 mg/mL (rat)
Compound X metabolite B NTCP alpha constant for inhibition	1e10 (competitive)
Compound X metabolite B NTCP switch	1 (competitive)
Compound X metabolite B basolateral inhibition constant	4.17E-03 mg/mL
Compound X metabolite B basolateral alpha constant for inhibition	1 (non-competitive)
Compound X metabolite B basolateral switch	0 (non-competitive)

[†]Funk 2001, Dawson 2011, [‡]Marion 2007, [§]Yang 2014

Preclinical Data



Institute for Drug Safety Sciences



CONFIDENTIAL

Defining Drug Toxicity Parameters in DILIsym[®]: Human Troglitazone BA Inhibition

DILIsym [®] Parameter	DILIsym [®] Parameter Input
Compound X BSEP inhibition constant	5.74E-04 mg/mL
Compound X BSEP alpha constant for inhibition	1e10 (competitive)
Compound X BSEP switch	1 (competitive)
Compound X NTCP inhibition constant	1.46E-04 mg/mL (human)
Compound X NTCP alpha constant for inhibition	1e10 (competitive)
Compound X NTCP switch	1 (competitive)
Compound X basolateral inhibition constant	9.27E-03 mg/mL
Compound X basolateral alpha constant for inhibition	1 (non-competitive)
Compound X basolateral switch	0 (non-competitive)

DILIsym Parameter Customization

Molecule: All Molecules | Mechanisms: All Mechanisms

Molecule	Mechanism	Variable	Value	Units
CompX	inhBAttransport	Compound X NTCP inhibition constant	1.4600e-04	mg/mL
		Compound X NTCP alpha constant for inhibition	1.0000e+10	dimensionless
		Compound X NTCP switch	1	dimensionless
		Compound X BSEP inhibition constant	5.7400e-04	mg/mL
		Compound X BSEP alpha constant for inhibition	1.0000e+10	dimensionless
		Compound X BSEP switch	1	dimensionless
		Compound X basolateral inhibition constant	0.0093	mg/mL
		Compound X basolateral alpha constant for inhibition	1	dimensionless
		Compound X basolateral switch	0	dimensionless
		Compound X ASBT inhibition constant	1.0000e+10	mg/mL

Panel View | Save w/ Custom | Cancel Changes | Save As New | Save As New w/ Custom

Simulating Troglitazone-Mediated Hepatotoxicity in Human SimPops™

HUMANS

Troglitazone 400mg/day for 6 months

DILIsym v4B

File Results View Help

SimSingle Setup

New SimSingle: Human_TGZ_400mg_6mo

Load SimSingle

Input Parameters

Species: Parameters_Species_Human_v4B

Drug: Parameters_Drug_Human_Troglitazone_v4B

Caloric Intake: Parameters_Calories_Human_v4B

Comp W Dosing: Parameters_CompWDosing_Blank_v4B

Comp X Dosing: 400mgperday_6months_oral

Comp Y Dosing: Parameters_CompYDosing_Human_v4B

Time: 6_months

Solver: Parameters_Solver_DeSolve_v4B

Input Panel: Panel_Blank

Simulate Run in Parallel **SimPops** Param Sweep Data Comparison

Specify Data

Plot Table Export Save Results SimSingle

SimPops™

Human_troglitazone_bile_acid_v3B_6

DILIsym SimPops

SimSingle Base File: Human_TGZ_400mg_6mo

SimPops File: Human_troglitazone_bile_acid_v3B_6

Sample size of SimPops (n): 331

Variables included in SimPops:

- Body_mass
- CDCA_amidation_Vmax
- CDCAamide_baso_Vmax
- CDCAamide_canal_Vmax
- CDCAamide_uptake_Vmax
- CompX_Met_B_bil_cl
- CompX_Vmax_L_B
- Km_CDCAamide_tox_direct
- Km_LCAsulfate_tox_direct
- LCA_synthesis_Vmax
- LCAamide_sulfation_Vmax
- LCAamide_sulfation_Vmax

Load initial conditions for SimPops:

Select DILIsym Outputs: New output panel All

Save SimPops results to data file:

Run Cancel

Exploring Troglitazone-Mediated Hepatotoxicity in Human SimPops™

Load SimPops™ Results
Tro_400mg_6mo_v3B6

HUMANS

The screenshot shows the 'SimSingle Setup' window with various input parameters. At the bottom, the 'Table' button is highlighted with a red circle. A red arrow points from this button to the 'DILIsym v4B Output Table' window.

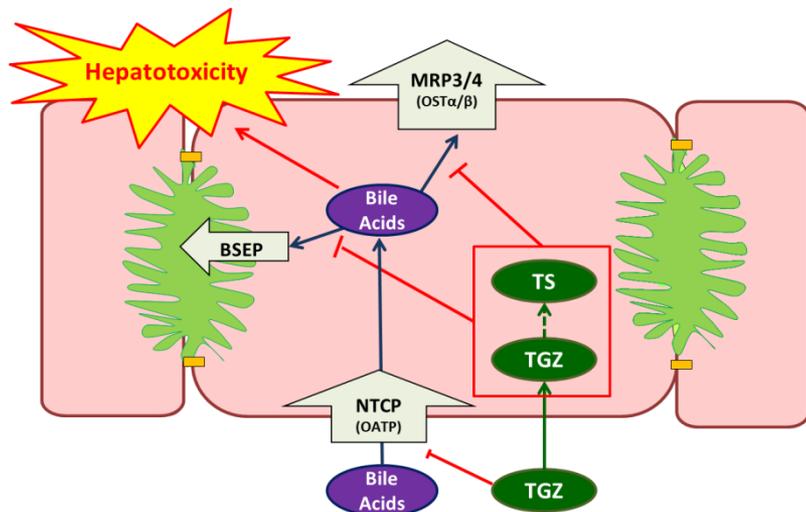
Group	Subgroup	Output Variable	Metric	Value	Units
All Groups	All Subgroups	None Selected	None Selected		
Outcomes	Outcomes	Number of deaths	Count		1 dimensionless
Outcomes	Outcomes	ALT at or over 3x ULN	Count		10 dimensionless
Outcomes	Outcomes	Bilirubin over 2x ULN	Count		6 dimensionless
	Outcomes	Hy's Law cases	Count		6 dimensionless

Buttons: Calculate, Reset

Troglitazone 400mg/day for 6 months

Output	Incidence
Number of deaths	1 / 331 (0.3%)
ALT elevations > 3X	10 / 331 (3.0%)
Bilirubin elevations > 2X	6 / 331 (1.8 %)
Hy's Law cases	6 / 331 (1.8 %)

What is the Contribution of Troglitazone Sulfate to the Hepatotoxicity?



- Troglitazone sulfate is a more potent BSEP inhibitor compared to troglitazone
- Systemic/hepatic exposure to troglitazone sulfate is greater than troglitazone

DILIsym [®] Parameter	DILIsym [®] Parameter Input	Adjusted DILIsym [®] Parameter Input
Compound X metabolite B BSEP inhibition constant	1.20E-04 mg/mL	1.00E+10 mg/mL
Compound X metabolite B NTCP inhibition constant	1.46E-04 mg/mL (human); 1.02E-03 mg/mL (rat)	1.00E+10 mg/mL
Compound X metabolite B basolateral inhibition constant	4.17E-03 mg/mL	1.00E+10 mg/mL

Turning Off Troglitazone Sulfate BA Inhibition

DILIsym Parameter Customization

Molecule: All Molecules Mechanisms: All Mechanisms

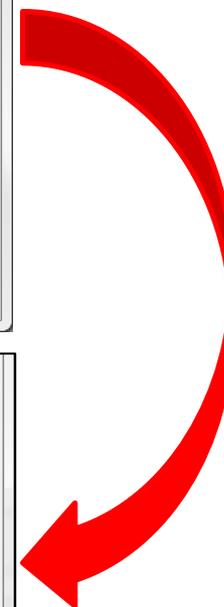
Molecule	Mechanism	Variable	Value	Units	
		Compound X ASBT inhibition constant	1.0000e+10	mg/mL	This par...
CompXMetB	inhBAtransport	Compound X metabolite B NTCP inhibition constant	1.4600e-04	mg/mL	This par...
		Compound X metabolite B NTCP alpha constant for inhibition	1.0000e+10	dimensionless	This par...
		Compound X metabolite B NTCP switch	1	dimensionless	0 = nonco...
		Compound X metabolite B BSEP inhibition constant	1.2000e-04	mg/mL	This par...
		Compound X metabolite B BSEP alpha constant for inhibition	1.0000e+10	dimensionless	This par...
		Compound X metabolite B BSEP switch	1	dimensionless	0 = nonco...
		Compound X metabolite B basolateral inhibition constant	0.0042	mg/mL	This par...
		Compound X metabolite B basolateral alpha constant for inhibition	1	dimensionless	This par...
		Compound X metabolite B basolateral switch	0	dimensionless	0 = nonco...

Panel View Save w/ Custom Cancel Changes Save As New Save As New w/ Custom

Molecule: All Molecules Mechanisms: All Mechanisms

Molecule	Mechanism	Variable	Value	Units	
		Compound X ASBT inhibition constant	1.0000e+10	mg/mL	This parameter represents the apical...
CompXMetB	inhBAtransport	Compound X metabolite B NTCP	1.0000e+10	mg/mL	This parameter represents the sod...
		Compound X metabolite B NTCP alpha co...	1.0000e+10	dimensionless	This parameter represents the sodi...
		Compound X metabolite B NTCP switch	1	dimensionless	0 = noncompetitive/mixed, 1 = compe...
		Compound X metabolite B BSEP	1.0000e+10	mg/mL	This parameter represents the bile...
		Compound X metabolite B BSEP alpha co...	1.0000e+10	dimensionless	This parameter represents the bile sal...
		Compound X metabolite B BSEP switch	1	dimensionless	0 = noncompetitive/mixed, 1 = compe...
		Compound X metabolite B basolateral	1.0000e+10	mg/mL	This parameter represents the bas...
		Compound X metabolite B basolateral alph...	1	dimensionless	This parameter represents the basolat...
		Compound X metabolite B basolateral swit...	0	dimensionless	0 = noncompetitive/mixed, 1 = compe...

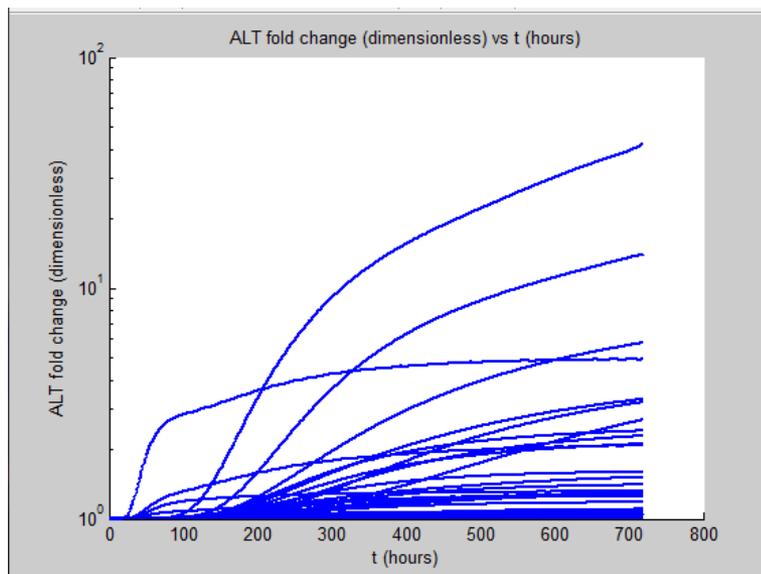
Panel View Save w/ Custom Cancel Changes Save As New Save As New w/ Custom



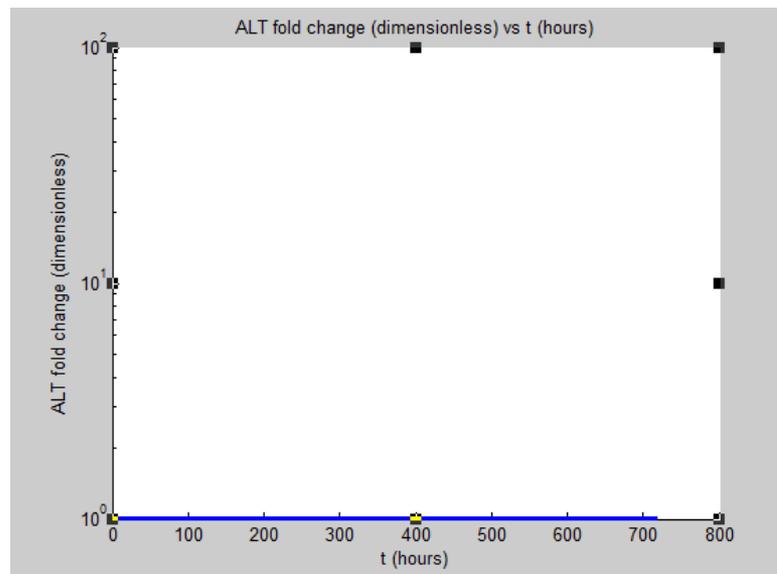
Troglitazone Sulfate is an Important Contributor for Hepatotoxicity

HUMANS

Troglitazone 400mg/day for 1 months



Troglitazone 400mg/day for 1 months – No troglitazone sulfate effects



Output	Incidence
Number of deaths	0 / 331 (0%)
ALT elevations > 3X	6 / 331 (1.8%)
Bilirubin elevations > 2X	1 / 331 (0.3%)
Hy's Law cases	1 / 331 (0.3%)

Output	Incidence
Number of deaths	0 / 331 (0%)
ALT elevations > 3X	0 / 331 (0%)
Bilirubin elevations > 2X	0 / 331 (0%)
Hy's Law cases	0 / 331 (0%)