Sensitivity analysis of P-glycoprotein Ki values in dynamic DDI predictions

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Background

The advantages of using PBPK models for prediction of transporter-mediated DDIs have been recognised [1]; although at present the observed degree of interaction is often under-predicted. One of the potential issues is the large variability in measured IC_{50} values [2]. The importance of using sensitivity analysis for key experimentally determined parameters has been highlighted in recent draft guidance for PBPK modelling [3,4]. The objective of this work was to investigate the fold range of intestinal P-glycoprotein (P-gp) Ki values required to recover digoxin DDIs with known inhibitors.

Methods

Published clinical studies involving inhibition of intestinal P-gp, using oral digoxin as the victim compound, were identified using the University of Washington drug interaction database [5].

In vitro P-gp inhibition data (IC_{50}) for perpetrator compounds, measured in Caco-2 cells with digoxin as the probe substrate, were collated from the literature. IC_{50} values determined using the efflux ratio (ER) approach (equation 1) were favoured; if only net secretory flux (NSF; equation 2) or unidirectional flux (UF; equation 3) approaches had been used the data were corrected to representative ER values (ER values are on average 3-fold lower than NSF or UF [2]).

$$ER = \frac{[(BAi/ABi) - (BAp/ABp)]}{[(BA0/AB0) - (BAp/ABp)]} \quad (equation\ 1)$$

$$NSF = \frac{(ABi - BAi)}{(AB0 - BA0)} \quad (equation\ 2) \qquad UF = \frac{(BAi - BAp)}{(BA0 - BAp)} \quad (equation\ 3)$$

DDI simulations were performed (Simcyp Simulator V15.1) using the clinical study designs and the default Simcyp library file for digoxin. For perpetrator compounds, the default Simcyp library files were modified to include P-gp Ki values calculated from IC_{50} data using the Cheng-Prusoff equation [6].

Sensitivity analyses for Ki were used to determine the values required to recover the observed *in vivo* C_{max} ratios.

Results

Healthy volunteer DDI studies with orally administered digoxin as the victim compound were identified for clarithromycin [7], itraconazole [8], ritonavir [9] and verapamil [10]. C_{max} ratios for digoxin in the presence of each of the four compounds were 1.83, 1.34, 1.26 and 1.44, respectively.

A range of IC_{50} values were identified for each compound (table 1).

Table 1: *In vitro* IC_{50} values for P-gp inhibitors clarithromycin, itraconazole, ritonavir and verapamil determined in Caco-2 cells with digoxin as the substrate via ER, UF and NSF methods

Inhibitor	IC_{50} [µM] ER	UF	NSF	Digoxin [µM]	Reference
Clarithromycin	*1.37**		4.1	5	[11]
	7	17		1	[12]
	34		66	5	[13]
Itraconazole	0.46	0.83		1	[12]
	0.69	0.38	0.862	5	[14]
	2		6	5	[13]
Ritonavir	*1.27**		3.8	5	[15]
	1.5	4.5		1	[12]
	5		10	5	[13]
Verapamil	0.2	0.5	0.7	10	[2] Lab 18
	*0.367**		1.1	5	[16]
	0.5	1.6	2.2	0.013	[2] Lab 16
	*0.7**		2.1	5	[15]
	0.9		1.2	1	[2] Lab 17
	*1.33**	4		5	[17]
	4		10	5	[13]
	4.9	25.8	48.3	1	[2] Lab 12
	*5.6**	16.8		0.1	[18]
	6.9	7.1	25	5	[2] Lab 14
	9.5	10	14.8	5	[2] Lab 8

*calculated from UF value/3 or NSF value/3

Simulations using Ki values calculated from the lowest IC_{50} (ER method) for each compound (table 2) were unable to recover the *in vivo* C_{max} ratios.

The results of the sensitivity analyses (figure 1) revealed that Ki values of <0.1 µM were required for all four compounds.

The difference between the 'fitted' and *in vitro* Ki values (table 2) ranged from 4.1-fold to 654-fold, with a mean of 94-fold. Considering only the lowest value for each compound, the mean fold difference was 19.5-fold.

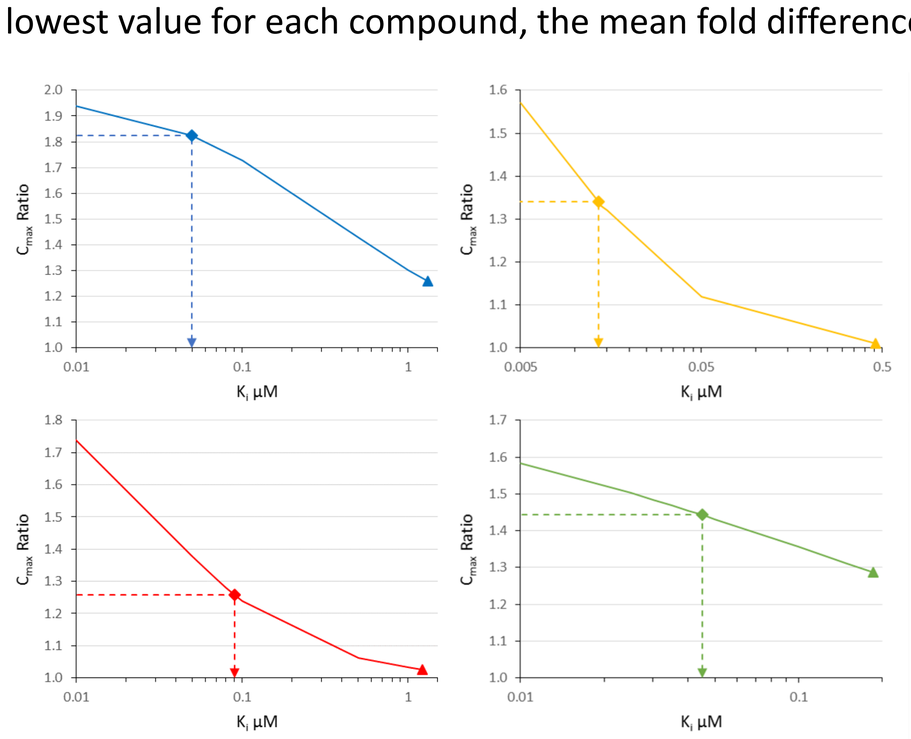

Figure 1: Sensitivity analyses for the impact of P-gp Ki values for clarithromycin (blue), itraconazole (yellow), ritonavir (red) and verapamil (green) on the prediction of *in vivo* digoxin C_{max} ratios

Table 2: The fold difference between the *in vitro* and 'fitted' P-gp Ki values for clarithromycin, itraconazole, ritonavir and verapamil

Inhibitor	IC_{50} [µM]	Ki [µM]	Fitted Ki [µM]	Difference (fold)
Clarithromycin	*1.37*	1.32	0.05	26
	7	6.96		139
	34	32.7		654
Itraconazole	0.46	0.457	0.0135	34
	0.69	0.671		50
	2	1.95		144
Ritonavir	*1.27*	1.22	0.09	14
	1.5	1.49		17
	5	4.81		53
Verapamil	0.2	0.185	0.045	4.1
	0.367	0.353		7.8
	0.5	0.5		11
	0.7	0.673		15
	0.9	0.893		20
	1.33	1.29		29
	4	3.85		86
	4.9	4.86		108
	5.6	5.6		124
	6.9	6.64		148
	9.5	9.14		203

Conclusions

- *In vitro* P-gp inhibition data required an average fold decrease of 94-fold (19.5-fold considering only lowest *in vitro* values for each inhibitor) to recover the *in vivo* interactions with digoxin.
- Potential reasons may relate to the (pre-)incubation conditions, inhibitor binding in the assay and inhibitory metabolites.

References

[1] Zamek-Gliszczynski MJ *et al.,* (2013) *Clin. Pharmacol. Ther.* 94(1): 64-79
[2] Bentz J *et al.,* (2013) *Drug Metab. Dispos.* 41(7): 1347-66
[3] EMA CHMP (2016) Guideline on the qualification and reporting of physiologically based pharmacokinetic (PBPK) modelling and simulation
[4] U.S. FDA CDER (2016) Physiologically Based Pharmacokinetic Analyses —Format and Content
[5] https://didb.druginteractioninfo.org, accessed September 2016
[6] Cheng, Y.-Ch. and Prusoff, W. H. (1973) *Biochem. Pharmacol.* 22:3099-3108
[7] Rengelshausen J *et al.*, (2003) *Br. J. Clin. Pharmacol.* 56: 32-38
[8] Jalava K-M *et al.*, (1997) *Ther. Drug Monit.* 19(6): 609-613
[9] Penzak SR *et al.*, (2004) *Ther. Drug Monit.* 26: 322-330
[10] Rodin SM *et al.*, (1988) *Clin. Pharmacol. Ther.* 43: 668-72
[11] Eberl S *et al.*, (2007) *Clin. Pharmacokinet.* 46 (12): 1039-1049
[12] Kishimoto W *et al.*, (2013) *Drug Metab. Dispos.* 42: 257-63
[13] Cook JA *et al.*, (2009) *Mol. Pharmaceutics* 7 (2): 398-411
[14] Volpe DA *et al.*, (2013) *AAPS J.* 16(1): 172-80
[15] Choo EF *et al.*, (2000) *Drug Metab. Dispos.* 28: 655–660
[16] Pauli-Magnus C *et al.*, (2000) *J. Pharmacol. Exp. Ther.* 293: 376–382
[17] Elsby R *et al.*, (2008) *Xenobiotica* 38(7–8): 1140–1164
[18] Kawahara I *et al.*, (2000) *Drug Metab. Dispos.* 28: 1238–1243