**BACKGROUND**

Accurate prediction of *in vivo* clearance is required for understanding drug efficacy and toxicity during drug development. Howgate *et al.* (2006) successfully predicted in vivo clearance for 25 drugs, in contrast to other studies noting an *under-prediction* trend (Houston et al., 1997; Obach et al., 1997 and 1999; Hallifax *et al.*, 2010). Traditionally, methods involve human liver microsomal (HLM) or hepatocyte (HHEP) data, using ‘average human’ scaling factors and comparing to one clinical study. However, CYP3A substrates have large *inter-individual variability* of *in vivo* clearance (Galetin *et al.*, 2004; Rawden *et al.*, 2005). Assessment of *inter-individual* variability of *in vivo* clearance allows analysis of range and identification of individuals with extreme clearance values.

**STUDY AIMS**

- **1) Clearance prediction accuracy** for alprazolam, triazolam and midazolam
- **2) Impact of variable IVIVE parameters** on variability of predicted *in vivo* CL
- **3) Variability in CYP3A enzyme abundance**: Separation of *inter-individual* from experimental variability

**METHODS**

1) **Bottom-up**: IVIVE of *in vitro* intrinsic clearance (CL\text{po}) and Top-down: Back-calculation from *in vivo* intravenous clearance (CL\text{iv})

*Figure 1. Incorporation of population-specific variability into predicted *in vivo* clearance using *in vitro* recombinant CYP (rhCYP) data*

Grey boxes: Incorporation of variability

- *i* = no. of metabolic pathways
- *j* = no. of CYP isoforms

2) **Simulations (Simcyp V10)**: Trial design mimicked clinical studies. Comparison to 10 randomly selected studies for each drug and both CL\text{po} & CL\text{iv} (except alprazolam and triazolam CL\text{po} only 4 studies available). Dataset of >150 studies. Variability inputs (% CV) were removed for parameters in turn (CYP3A4 liver/gut abundance, MPPGL, liver volume, haemocrit).

Impact on variability of CL\text{po} and CL\text{iv} was assessed.

3) Separation of *inter-individual* from experimental variability in CYP3A4 abundance using repeat measurement ELISA protocol in individual HLM (n=52).

**RESULTS**

1) **Clearance prediction accuracy**

Bottom-up: Predicted clearances were within 2-fold of observed for triazolam and midazolam but 2 to 3.7-fold higher than observed for alprazolam.

Top-down: *In vivo* CL\text{iv} allowed more accurate assessment of variability of *in vivo* clearance when predictions were optimal (within 2-fold) (Figure 2).

2) **Impact of parameter CV on variability in predicted triazolam CL**

The IVIVE parameters with the greatest impact on variability of predicted *in vivo* clearance were hepatic CYP3A4 abundance and MPPGL. As CV values for these parameters were increased from 0-100% in turn, variability of *in vivo* clearance increased by 230% (both CL\text{po}, and CL\text{iv}; hepatic CYP3A4 abundance) and 39% and 62% (CL\text{po} and CL\text{iv}, respectively; MPPGL).

3) **Separation of *inter-individual* from experimental variability for hepatic CYP3A44 abundance**

CV for hepatic CYP3A4 abundance from literature meta-analysis was 95%. CV for hepatic CYP3A4 abundance from experimental data (representing ‘true’ *inter-individual* variability - repeat measurement ELISA protocol) was 41%.

Large variability in observed clearance was seen between different clinical studies. Mean CL\text{po}: Ranged 1.4, 1.8 and 2-fold for alprazolam, triazolam and midazolam, respectively. Mean CLpo: Ranged 1.5, 2.5 and 3.3-fold, respectively.

**CONCLUSIONS**

- *In vitro* rhCYP data can be used to accurately predict *in vivo* clearance for a range of different clinical studies (seen here for triazolam and midazolam).
- Different clinical studies show significant variability of *in vivo* clearance.
- A lack of incorporation of variability in both *in vitro* and *in vivo* data could contribute to inconsistent accuracy of clearance predictions (Houston *et al.*, 1997; Obach *et al.*, 1997 and 1999; Howgate *et al.*, 2006; Hallifax *et al.*, 2010).
- There is a need for refinement of reported values of variability for IVIVE parameters (to distinguish experimental and *inter-individual* variability).
- Reduction of variability in hepatic CYP3A44 abundance to a value representing only *inter-individual* variability (CV 41%) would seem the best approach for estimation of variability of CYP3A44 abundance using *in vitro* elimination data.

**REFERENCES**


**Study accepted for publication. Available ‘Early Online’: http://informahealthcare.com/xen**