

Cloe Screen Cytochrome P450 Inhibition

Background Information



'The effects of new drugs on well characterized drug metabolism reactions known to be specific for various human drug-metabolizing enzymes are routinely examined using *in vitro* approaches.'

¹Obach RS, Walsky RL, Venkatakrisnan K, Gaman EA, Houston JB and Tremaine LM. (2006) *JPET* 316; 336-348.

- Cytochrome P450 are a family of enzymes which play a major role in the metabolism of drugs.
- Assessment of the potential of a compound to inhibit a specific cytochrome P450 enzyme is important as co-administration of compounds may result in one or both inhibiting the other's metabolism. This may affect plasma levels *in vivo* and potentially lead to adverse drug reactions or toxicity.
- *In vitro* cytochrome P450 inhibition data are useful in designing strategies for investigating clinical DDI Studies.
- Cloe Screen Cytochrome P450 Inhibition assays meets criteria set out in FDA guidelines.
- In the Cloe Screen Cytochrome P450 Inhibition assay, a decrease in the formation of the metabolites compared to the vehicle control is used to calculate an IC₅₀ value (test compound concentration which produces 50% inhibition).

Protocol

Test Compound Concentration
0, 0.1, 0.25, 1, 2.5, 10, 25 μ M
(different concentrations available)

CYP Isoforms
CYP1A, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP3A4

Compound Requirements
200 μ L of 10 mM solution

Controls
Known isoform specific inhibitors

Analysis method
LC-MS/MS

Data Delivery
IC₅₀
Standard error of IC₅₀

In vitro P450 inhibition data are valuable in the design of clinical DDI study strategies and can be used to predict the magnitudes of DDI¹.

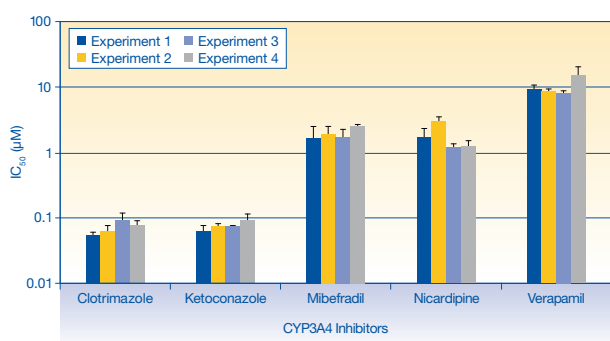


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Known cytochrome P450 inhibitors were screened in the Cloe Screen Cytochrome P450 Inhibition assay in quadruplicate over 4 separate assays.

Figure 1

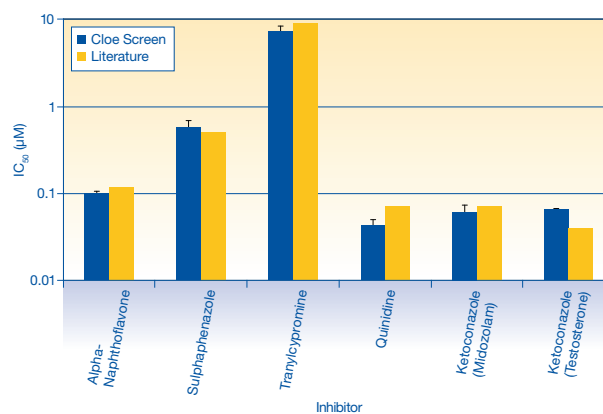
Cloe Screen Cytochrome P450 Inhibition data for CYP3A4.



The effect of 5 known CYP3A4 inhibitors (clotrimazole, ketoconazole, mibefradil, nicardipine and verapamil) on the 1-hydroxylation of midazolam was investigated on 4 separate occasions. Error bars represent the standard deviation of 4 replicates on each experiment. The data show good consistency for inhibitors with a range of inhibition potential.

Figure 2

Comparison of Cloe Screen IC_{50} values (mean \pm standard deviation) for the control inhibitors with literature^(2,3,4,5,6,7) values.



References

- Obach RS *et al.* (2006) *JPET* **316**; 336-348.
- Bu HZ *et al.* (2001) *Eur J Pharm Sci* **12** (4); 447-52.
- Back DJ *et al.* (1988) *Br J Clin Pharmacol* **26** (1); 23-29.
- Dierks EA *et al.* (2001) *Drug Metab Dispos* **29** (1); 23-9.
- Eagling VA *et al.* (1998) *Br J Clin Pharmacol* **45** (2); 107-114.
- Moody GC *et al.* (1999) *Xenobiotica* **29** (1); 53-75.
- Nomeir AA *et al.* (2001) *Drug Metab Dispos* **29** (5); 748-53.