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In vitro **ADME & PK**

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.'

[1]Obach RS. Walsky RL, Venkatakrishnan K, Gaman EA, Houston JB and Tremain 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.
- Cyprotex's Cytochrome P450 Inhibition assays use industry accepted probe substrates and human liver microsomes.
- In Cyprotex's Cytochrome P450 Inhibition assay, a decrease in the formation of the metabolites compared to the vehicle control is used to calculate an IC_{50} value (test compound concentration which produces 50% inhibition).

Protocol

Typical Test Article Concentrations
0, 0.1, 0.25, 1, 2.5, 10, 25 µM
(different concentrations available)

CYP Isoforms
CYP1A, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6 and CYP3A4
(other isoforms are available)

Test Article Requirements
Dependent on number of isoforms assessed

Controls
Known isoform specific inhibitors

Analysis Method
LC-MS/MS (with the exception of ethoxyresorufin for CYP1A)

Data Delivery
IC_{50}
Standard error of IC_{50}

To find out more contact **enquiries@cyprotex.com**

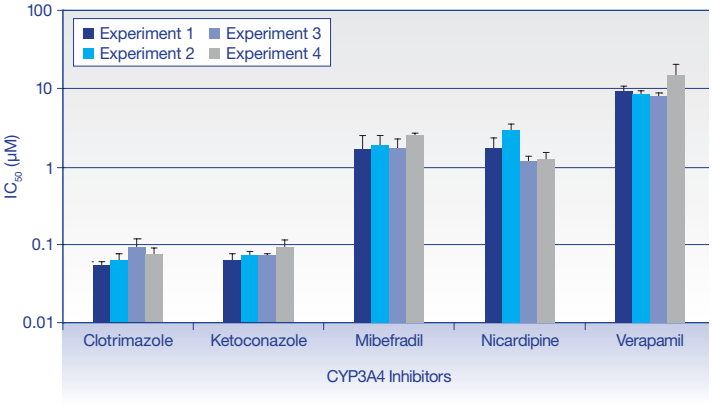
***In vitro* P450 inhibition data** are valuable in the design of clinical DDI study strategies and can be used to predict the magnitudes of DDI[1].

Cytochrome P450 Inhibition

Known cytochrome P450 inhibitors were screened in Cyprotex's Cytochrome P450 Inhibition assay in quadruplicate over 4 separate assays.

Figure 1

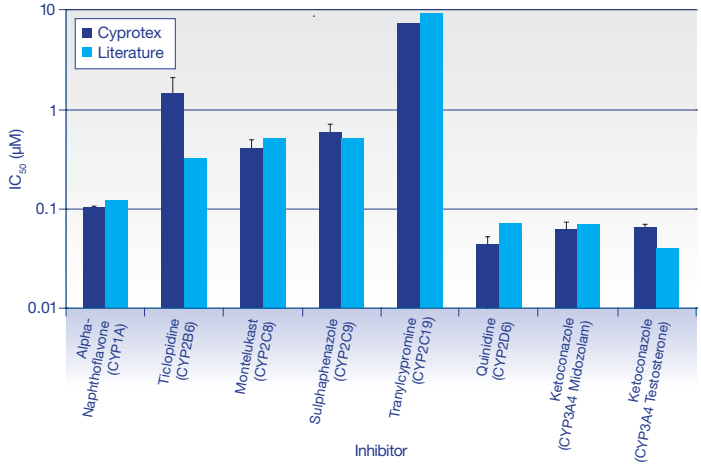
Cyprotex's Cytochrome P450 Inhibition data for CYP3A4.



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 Cyprotex's IC_{50} values (mean± standard deviation) for the control inhibitors with literature[2,3,4,5,6,7,8] values.



References

1 Obach RS *et al.* (2006) *JPET* **316**; 336-348.
2 Bu HZ *et al.* (2001) Eur *J Pharm Sci* **12 (4)**; 447-52.
3 Turpeinen M *et al.* (2004) *Drug Metab Dispos* **32 (6)**; 626-631.
4 Back DJ *et al.* (1988) *Br J Clin Pharmacol* **26 (1)**; 23-29.
5 Dierks EA *et al.* (2001) *Drug Metab Dispos* **29 (1)**; 23-9.
6 Eagling VA *et al.* (1998) *Br J Clin Pharmacol* **45 (2)**; 107-114.
7 Moody GC *et al.* (1999) *Xenobiotica* **29 (1)**; 53-75.
8 Nomeir AA *et al.* (2001) *Drug Metab Dispos* **29 (5)**; 748-53.