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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**

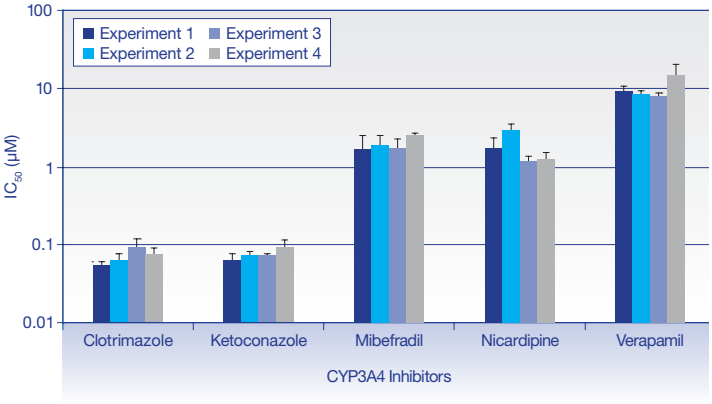
***In vitro* P450 inhibition data** are valuable in the design of clinical DDI study strategies and can be used to predict the magnitudes of DDI[1].

## Cytochrome P450 Inhibition

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

**Figure 1**

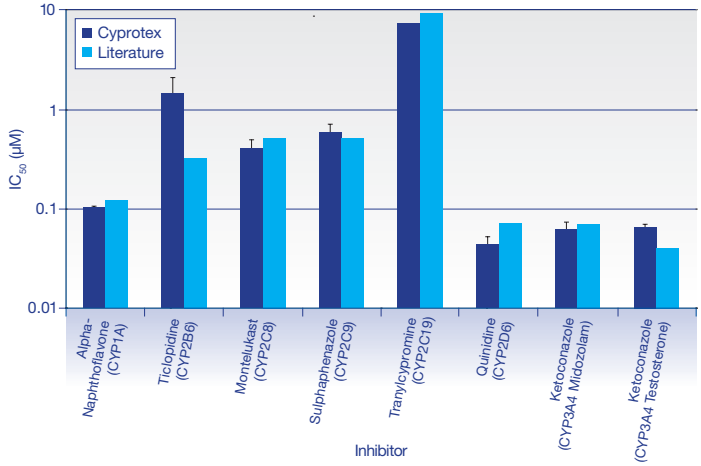
Cyprotex's Cytochrome P450 Inhibition data for CYP3A4.




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.