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In vitro **ADME & PK**

Aldehyde Oxidase (AO) Reaction Phenotyping

Background Information

'It has been increasingly recognized in this past decade that AO, through its unique structure, distribution, and substrate recognition, has an important role to play in the metabolism of drugs.'

[1]Pryde DC *et al*. (2010), *J. Med. Chem*., **53**; 8441-8460

- Aldehyde oxidase is a cytosolic enzyme and is a sub family of the molybdo-flavoenzymes.
- For catalytic activity, aldehyde oxidase requires a molybdo-pterin cofactor (molybdenum cofactor, MoCo) and flavin adenine dinucleotide.
- It has broad substrate specificity and catalyses the oxidation of aldehydes into carboxylic acids and also hydroxylation of some N-heterocycles.
- The main isoform in humans is AO1 which has highest activity in the liver. The enzyme has also been detected in other tissues such as the lung, gastrointestinal tract and kidney.
- Cyprotex's aldehyde oxidase (AO) reaction phenotyping assay determines if your compound is a substrate for aldehyde oxidase (AO).

Protocol

Test System
Human liver cytosol incubated with and without specific AO inhibitor, 100 µM menadione (other species and enzyme sources available on request)

Test Article Concentration
1 µM (different concentrations available)

Positive Control Substrate
Phthalazine

Time Points
0, 10, 20, 40, 60, 120 min

Test Article Requirements
100 µL of a 10 mM DMSO solution (or equivalent amount in solid)

Analysis Method
LC-MS/MS

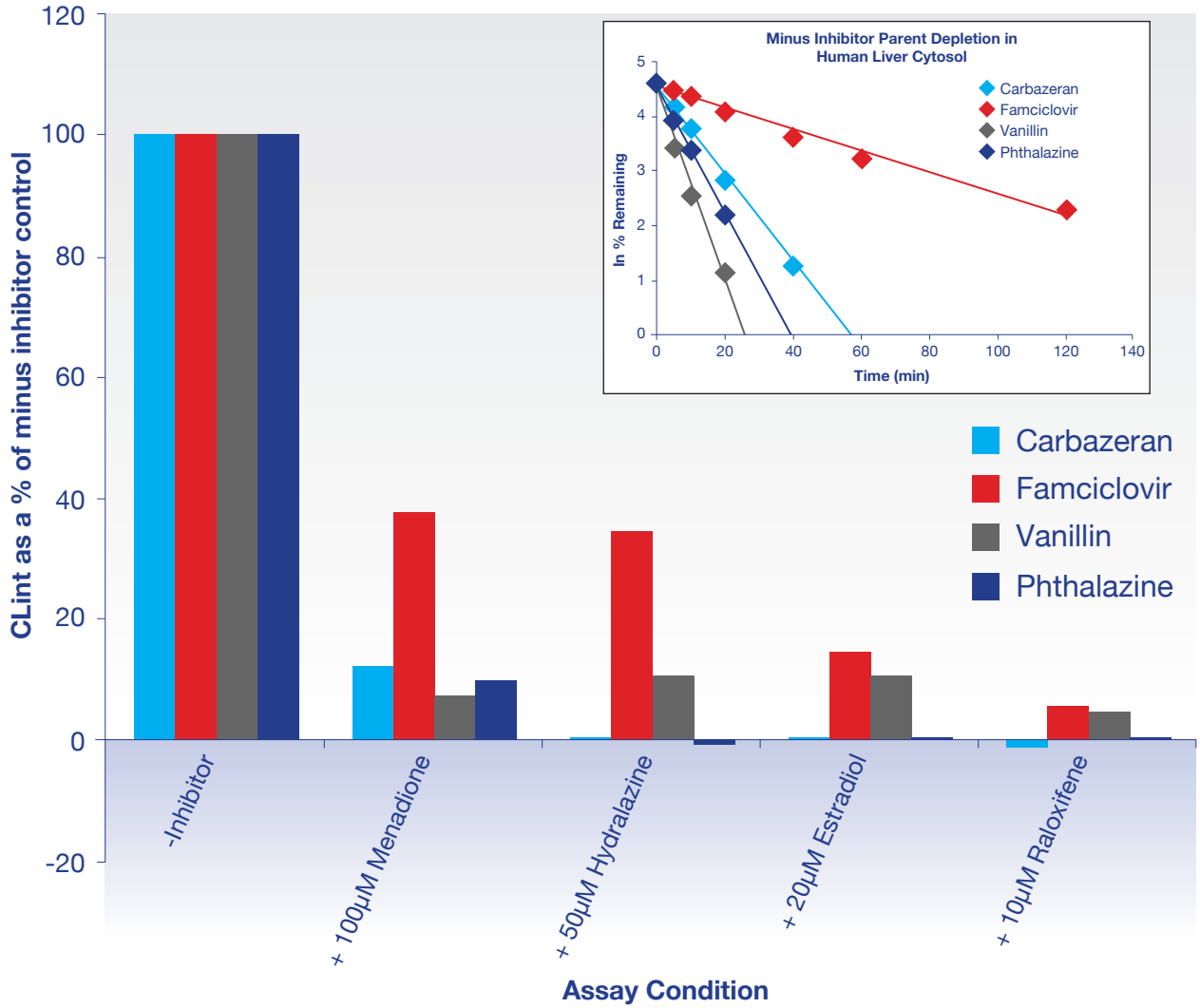
Data Delivery
% Parent compound remaining at each time point
Intrinsic Clearance
Half life
Standard error of half life

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

'AOX-dependent biotransformation of new drug candidates is an emerging problem, as new strategies of chemical synthesis aimed at reducing CYP450-dependent metabolism tend to enrich for pharmacophores, which are AOX substrates and are inactivated by this enzymatic system. This calls for the development of new approaches to predict and test AOX-dependent metabolism particularly.'[2]

Figure 1

Results from human liver cytosol stability of AO substrates carbazeran, famciclovir, vanillin and phthalazine in the presence of different AO inhibitors, displayed as a percentage of the CLint of the minus inhibitor control compound. Inset: displays substrate depletion over time in the absence of inhibitor.



References

[1] Pryde DC *et al*. (2010) Aldehyde oxidase: an enzyme of emerging importance in drug discovery. *J Med Chem* **53**; 8441-8460

[2] Garattini E & Terao M (2013) Aldehyde oxidase and its importance in novel drug discovery: present and future challenges. *Expert Opin Drug Discov* **8(6)**; 641-654