POLYCYPS: BIOCATALYTIC TOOLS FOR THE OPTIMISATION OF DRUG LEADS

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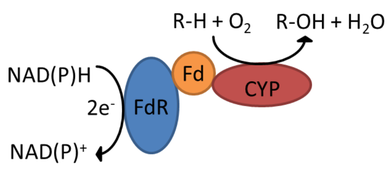
Abstract This poster illustrates the application of PolyCYPs biocatalysts to generate hydroxylated and other oxidised derivatives that complement conventional lead optimisation strategies. PolyCYPs are recombinant cytochrome P450 enzymes cloned from talented bacteria in Hypha's biotransformation panel. The production of multiple hydroxylated derivatives in parallel by PolyCYPs gives the opportunity to identify derivatives with improved properties.

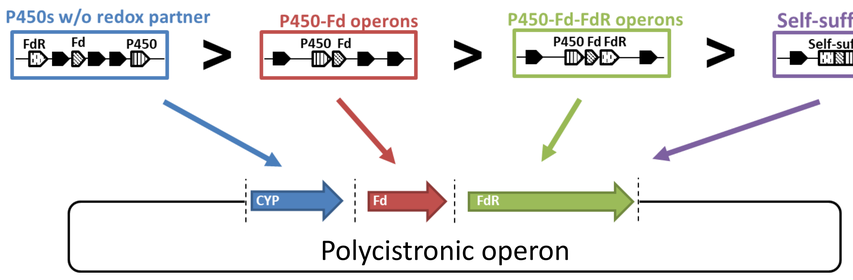
Potential benefits

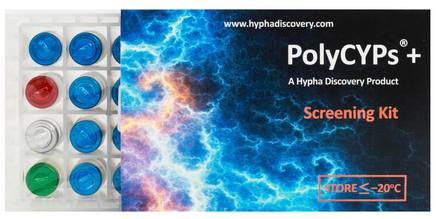
- Boost DMPK properties, particularly metabolic stability
- Empirically discover new polar interactions in binding sites, improving potency and selectivity
- Establish if metabolites are active before deprioritising a metabolically unstable scaffold
- Utilise hydroxylated derivatives/metabolites as handles for late stage functionalisation e.g. fluorination
- Rapidly expand polar SAR and broaden IP coverage, including the exemplification of active metabolites

About PolyCYPs

PolyCYPs® enzymes are class I cytochrome P450s mined from selected bacteria in Hypha's strain collection and cloned into a polycistronic operon with native or surrogate redox partners. They are expressed in both *E.coli* and *Streptomyces lividans* hosts and catalyse a broad range of oxidative reactions mimicking the metabolising capability of human CYPs.







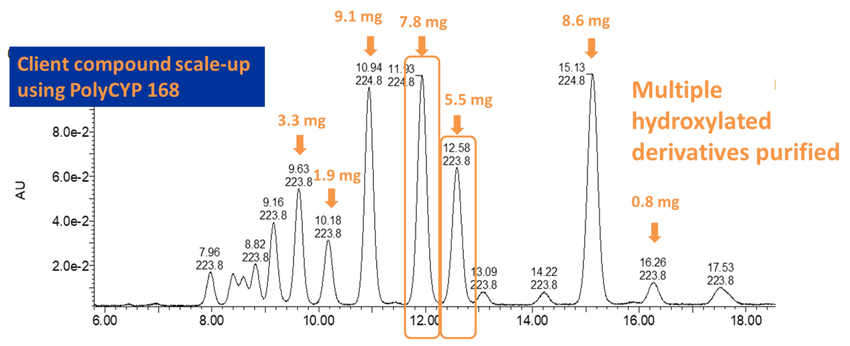
PolyCYPs enzymes are available in screening and scale-up kit form as lyophilized cell-free extracts. There are 18 PolyCYP isoforms in the screening kit together with human recombinant aldehyde oxidase and flavin monooxygenase 3.

As well as generating metabolites, PolyCYPs enzymes can be used to create oxidised derivatives of drug compounds that may have improved properties compared to the parent drug. We call this process PolarExplorer™.

PolarExplorer process

Structure elucidation by NMR spectroscopy

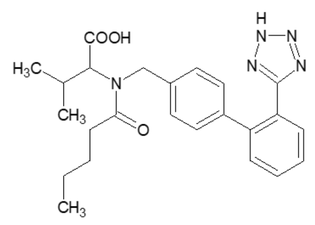
Sample chromatogram illustrating the parallel production of multiple hydroxylated derivatives by PolyCYPs



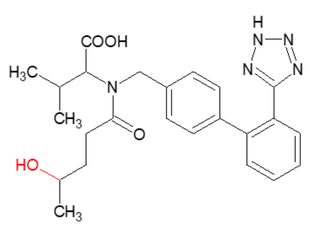
Two of the hydroxylated compounds identified showed interesting activity and were selected for further profiling by the client.

PolyCYPs in action - broadening IP

Valsartan case study



CYP2C9
Multiple PolyCYPs

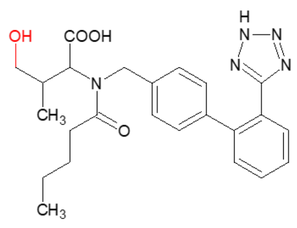


4-hydroxyvaleryl valsartan

Possesses novel ARB-independent platelet aggregation activity

Patented by Novartis

PolyCYP 168



Novel hydroxylated derivative

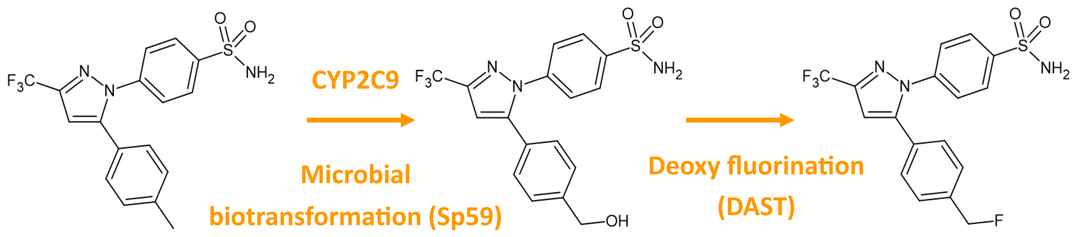
Angiotensin receptor blocker (Novartis drug)

Minimally metabolised in HLMs by CYP2C9 to 4-hydroxyvaleryl valsartan

PolyCYPs used to quickly generate metabolites and oxidised derivatives with enhanced properties that could widen IP coverage.

PolyCYPs in action - providing synthetic handles

Celecoxib case study



- Fluorination is a well known strategy for improving metabolic stability of drugs. A late-stage hydroxylation-fluorination strategy can be used to test this.
- 4'-fluorocelecoxib is 4 fold more metabolically stable to CYP2C9 metabolism but is metabolised faster in human liver microsomes, possibly due to metabolism by other CYP enzymes.[1]

This methodology can be used to test the impact of fluorination in modulating CYP metabolism, particularly for those CYPs known to be implicated in drug-drug interactions or subject to genetic polymorphism.

Reference: [1]Obach et al., 2016. Drug Metab Dispos 44:634–646.

About Hypha: Hypha Discovery is a specialist CRO with expertise in the scalable synthesis, purification and identification of drug metabolites. Using our biocatalytic and organic chemistry capabilities, we also make oxidized derivatives of lead compounds and API degradation products. Hypha also has a wealth of experience in the production, purification and structure elucidation of natural products. We work with pharmaceutical and agrochemical companies globally.

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