Hepatic Microsomal Stability (human, rat or mouse)

Background: Liver is a primary site of drug metabolism, and drug metabolic transformations may have significant impact on its efficacy and safety. For this reason, drug candidates are screened early in the discovery process for metabolic stability. Microsomes from human or animal liver are useful models to quickly and inexpensively predict hepatic clearance in vitro for the corresponding species. Stability experiments can be done either with hepatic microsomal fraction to investigate only Phase I metabolism or with the S9 fraction, which consists of both hepatic microsomes and cytosol. The advantage of using S9 fraction is that it contains both Phase I and Phase II enzymes and can be used to investigate Phase II metabolic pathways in vitro, when supplemented with the corresponding cofactors such as UDPGA (for glucuronidation) and PAPS (for sulphation).

Service Details: Metabolic stability assays are typically performed using mouse, rat, or human microsomes or S9 fraction (microsomes from other species are available upon request). Test compounds are incubated with microsomes supplemented with cofactors at 37°C. Typical conditions are the compound concentration of 2 µM and 5 sampling time points over 40 min, in two independent replicates. At each time point, the reactions are terminated with acetonitrile. The samples are centrifuged and the relative parent compound depletion is evaluated by LC-MS/MS. The incubation of two control drugs with microsomes and blank control reaction without co-factors are used as controls.

Deliverable: Data include parent compound percent remaining, half-life ($t_{1/2}$), and intrinsic clearance (Cl_{int}) values. Full study report is provided.

Sample Submission: A minimal accurately weighable quantity of dry compound (~1 mg or 2 µmol) or 50 µL of 10-20 mM stock DMSO solution is required for this assay. For multiple assays, lesser amount of compound per assay may be sufficient, depending on the particular project. We do not need to know structures of the molecules for ADME testing. However, brutto formulas have to be provided for all studies involving MS detection.