MONASH University

CENTRE FOR DRUG CANDIDATE OPTIMISATION

BINDING AND PARTITIONING

An understanding of the extent to which a drug partitions into red blood cells and binds to proteins in plasma, tissues and in vitro assay media, allows for potentially improved in vitro-in vivo correlations of biological activity.

Binding of a compound to plasma proteins (albumin, α_1-acid glycoprotein or lipoproteins) or partitioning into erythrocytes can reduce the concentrations of free drug available for distribution from the circulation into tissues and sites of pharmacological activity. Binding can also reduce access of free drug to clearance mechanisms in the kidney and liver and therefore may reduce clearance of a compound from the body.

Knowledge of a compound's binding and partitioning properties can therefore help with the interpretation of in vivo pharmacokinetic and pharmacodynamic results. Binding of test compounds to plasma as well as tissues (e.g. brain) and media used for assessing biological activity can be useful for correlating unbound concentrations across in vitro and in vivo test systems and improving predictions of in vivo disposition in pharmacokinetic modelling.

The Centre for Drug Candidate Optimisation (CDCO) offers a range of assays to assess the binding of test compounds in various matrices including plasma, tissues, assay media or microsomes. Binding is experimentlaly determined using either rapid equilibrium dialysis (RED) or ultracentrifugation (UC) methods. Binding to plasma proteins from different species can be determined using either neat or diluted plasma, with diluted plasma providing better resolution of the unbound fraction for highly bound compounds.

Research platforms include:

- Fraction unbound (f_u) in plasma using either rapid equilibrium dialysis (RED) or ultracentrifugation
- Fraction unbound (f_u) determination in selected tissues, assay media and liver microsomes
- Whole blood-to-plasma partitioning ratio
- Chromatographic estimation of plasma protein binding: human serum albumin (HSA) and α_1-acid glycoprotein (AGP)

Available species include human, rat and mouse (check availability for dog and non-human primate).

Binding to HSA can be estimated using an HPLC method where the retention of a series of test compounds on immobilised HSA protein can be compared to the retention of a series of compounds with known protein binding values. This platform affords a medium-throughput method for screening and ranking a series of compounds. An AGP column can be included to provide a qualtitative assessment of likely binding to AGP.

The extent of partitioning of test compounds into erythrocytes can be determined in vitro or in vivo as part of a pharmacokinetic experiment.