Most Recent Previous projects in CAPKR

CAPKR CONSORTIUM FUNDED PROJECTS

Modelling endogenous biomarker of renal and hepatic transporters

The project aimed to develop mechanistic models for endogenous biomarkers of hepatic and renal transporters, to inform the optimal design and need for clinical transporter drug-drug interaction (DDI) studies in drug development. In addition, this project evaluated DDI risk in patients with renal impairment and investigated delineation of the effect of disease and drug-mediated inhibition of transporters in this patient population. Key outcomes included development of models for coproporphyrin I, creatinine and 4-pyridoxic acid to support evaluation of transporter DDI risk in early drug development. Extension of biomarker models to patients with renal impairment identified challenges in quantitative translation to patient populations, supporting the aim to use biomarkers as a novel tool to evaluate function of transporters *in vivo*.

PI/researchers: Amais Ahmad, Hiroyuki Takita, Dr Kayode Ogungbenro, Prof. Aleksandra Galetin

Funding: CAPKR

Optimising Co culture Systems to Improve Predictions of Low Hepatic Clearance

The project aimed to understand the impact of plasma binding in both limiting (free drug hypothesis) and enhancing (protein mediated uptake, PMU) hepatic clearance; to establish an assay that can generate reliable measurements of low clearance compounds in a dynamic hepatocyte environment; and assess the utility of the medium loss assay for co-culture systems to capture both transport and metabolic processes and inter-species (rat-human) applicability within the in vitro-in vivo extrapolation (IVIVE) context. A novel approach and incorporation of PMU is recommended to improve IVIVE and the challenges of investigating stable (low clearance, high protein binding) drugs with co-culture hepatocyte systems have been addressed.

PI/researchers: Prof. David Halifax, Prof. Brian Houston, Dr Kayode Ogungbenro, Dr.Laura Francis

Funding: CAPKR

Liquid Biopsy: Precision dosing of metabolically-cleared drugs- Development of techniques for precision dosing of metabolically-cleared drugs

The project aimed to evaluate liquid biopsy as an approach to achieve precision dosing in different patient cohorts by quantifying and assessing inter-individual variability of drug metabolising enzymes/transporters in plasma exosomes (based on previously established correlation of plasma read-out with tissue protein expression), in order to link them to established biomarkers of activity/phenotype; Integrating data into physiologically-based pharmacokinetic (PBPK) models to manage variable drug metabolism. The key conclusions from the study included liquid biopsy is a non-invasive, single-assay quantitative approach for monitoring inter-individual variability in metabolic enzymes for efficient patient characterization in diverse disease cohorts and that liquid biopsy-informed virtual twin models can guide precision dosing in patients with variable renal and hepatic function.

PI/researchers: Dr Brahim Achour, Professor Amin Rostami, Professor Aleksandra Galetin

Funding: CAPKR**; Industry collaborators**: Certara, Thermo, Illumina

Blood-brain barrier

The blood-brain barrier (BBB) remains a focal point of interest for many scientists who are working on approaches to deliver various therapeutic agents into the brain. This project aimed to refine methodologies for IVIVE-PBPK models to provide linkage from CSF to localized brain concentration for compounds with mild-to-moderate efflux liabilities. In vitro transporter kinetics experiments were performed for selected compounds in cell lines expressing relevant transporters of interest. Transporter kinetics, together with binding and permeability data were analysed with mechanistic model accounting for all these cellular processes. In addition, proteomics analysis of the expression of relevant transporters in the cell lines used for in vitro studies and tissues from the rat in vivo studies were performed in order to generate relevant scaling factors for translation purposes to *in vivo*. In addition to microdyalisis, a novel sampling technique based on conventional open flow microperfusion (cOFM) was explored for selected compounds. In house mechanistic model has been developed, but IVIVE PBPK work and verification against generated data still needs to be completed.

PI/researchers: Dr Kayode Ogungbenro, Dr Zubida Al-Majdoub, Professor Aleksandra Galetin, Professor Amin Rostami

Funding: CAPKR

Evaluating protein abundance vs. activity relationships of drug-metabolizing enzymes (CYP and UGT) in the human liver and small intestine

Current approaches for bottom-up prediction of intestinal and hepatic metabolic clearance, including biologically-associated variability, depend on assumption that enzyme activity is proportional to specific protein abundance. This activity-abundance proportionality is assumed to hold irrespective of the organ or patient characteristics. However, there is limited evidence to support this beyond the work done for intestinal and hepatic CYP3A4 (Gertz et al Drug Metab Dispos 2010, 38(7):1147-58).

The project investigated relationship between expression and functional activity of major CYP and UGT drug metabolizing enzymes between human liver and gut. Using CAPKR's expertise in functional and proteomic analysis, project aims to measure enzyme abundance and activity of specific probes in matched liver and intestinal samples obtained from the same human donors. Implications of the findings on the quantitative translation of drug clearance and drug-drug interactions will be investigated.

PIs/Researchers: Dr Zubida Al-Majdoub, Dr Aleksandra Galetin , Dr Jill Barber, Dr Daniel Scotcher, Professor Amin Rostami

Funding: CAPKR

Establishing a translational PBPK framework for human aldehyde oxidase)

The primary focus of the proposed project is to develop physiologically-based pharmacokinetic (PBPK) models for selected aldehyde oxidase (AO) substrates (and mixed aldehyde oxidase/ cytochrome P450 substrates). The PBPK models will be informed and verified by in vitro data, quantitative protein abundance and clinical pharmacokinetics and drug-drug interaction (DDI) data. The emphasis of the project is on elucidating the current gaps and limitations for quantitative translation of clearance and DDIs for AO substrate drugs within the PBPK framework. It is anticipated the research will lead to new approaches to delineate, quantify and predict the in vivo roles of hepatic and non-hepatic AO and other metabolic pathways (e.g., cytochrome P450). Expected outcome of the research will be a novel roadmap for in vitro-in vivo extrapolation linked with PBPK models for aldehyde oxidase-mediated metabolism and DDIs for application in drug discovery and development.

PIs/Researchers: Dr Daniel Scotcher, Prof Aleksandra Galetin, Dr Jill Barber, Prof Brian Houston, Dr Nihan Izat (postdoc)

Funding: CAPKR

NON-CAPKR FUNDED PROJECTS

Translational Imaging in Drug Safety Assessment

Imaging biomarkers, based on non-invasive techniques such as positron emission tomography (PET) and magnetic resonance imaging (MRI), offer potential for insights into toxicity issues early on in drug development. One of the aims is to validate the use of gadoxetate as MRI biomarker for assessment of hepatobiliary transporter mediated drug-drug interactions. CAPKR team performed in vitro evaluation of hepatic uptake of gadoxetate and develop a physiologically based pharmacokinetic model for this imaging biomarker. CAPKR's expertise in quantitative translation will support the mechanistic interpretation of the in vivo MRI data being generated for gadoxetate, including effects of various transporter inhibitors.

PI/researchers: Prof. Aleksandra Galetin (PI), working with Dr Adam Darwich, Dr Kayode Ogungbenro, and Dr Daniel Scotcher, Dr Nicola Mellilo (postdoc)

Funding: Innovative Medicines Initiative 2 Joint Undertaking under grant agreement No 116106

Evaluating drug-drug interaction risk associated with peptide analogues using advanced in vitro systems

The aim of the study was to investigate the ability of advanced human hepatocyte *in vitro* systems namely HepatoPac, spheroids, and Liver-on-a-chip to assess potential changes in regulation of CYP1A2, CYP2B6, CYP3A4, *SLCO1B1* and *ABCC2* in the presence of selected therapeutic peptides, proteins, and small molecules. HepatoPac, the most extensively investigated in this study (3 donors), showed high potential to investigate DDIs associated with CYP regulation by therapeutic peptides. Spheroids and Liver-on-a-chip were only assessed in one hepatocyte donor and further evaluations are required to confirm their potential. This study establishes an excellent foundation towards the establishment of more clinically-relevant *in vitro* tools for evaluation of potential DDIs with therapeutic peptides.

PIs/Researchers: CAPKR Prof Aleksandra Galetin; Novo Nordisk - Dr Carolina Säll, Dr Rune Nørgaard (postdoc)

Funding: Novo Nordisk

Improving Pharmacokinetics Prediction for Small Molecule Drugs using multi-organ microphysiological systems and translational modelling and simulation

The aim of the study was to investigate Liver-on-a-chip system for multiple drug metabolism applications. Characterization of CYPs, UGT and aldehyde oxidase (AO) activities was performed using 15 drugs, followed by quantitative extrapolation of generated data to *in vivo* . The utility of the liver-on-a-chip for estimation of the fraction metabolized (f_m) was also established. In addition, project explored microphysiological systems (MPS) consisting of gut-liver organ-on-a-chip components to investigate intestinal permeability, metabolism (intestinal and hepatic) and potential interplay of those processes using mycophenolate mofetil was tested for quantitative evaluation of the gut-liver organ-on-a-chip due to the contribution of both gut and liver in its metabolism. Both studies demonstrated the integration of mathematical modelling with experimental organ-on-a-chip studies and how this integrated approach can support generation of high quality of data from complex *in vitro* cellular systems.

PIs/Researchers: CAPKR Prof Aleksandra Galetin; Roche - Drs Michael Gertz, Stephen Fowler, Neil Parrott, Dr Nicolo Milani (postdoc)

Funding: Roche

***In vitro* screening of chemotherapeutic treatments and target therapies for Pseudomyxoma peritonei (PMP)**

The aim of the project was to profile the characteristics of the tumours in PMP patients and seek out specific drug targets for PMP. Tumour and histologically normal peri-carcinomateous tissues, as well as plasma and drain fluid collected from PMP patients undergoing cytoreductive surgery (CRS) and heated intraoperative peritoneal chemotherapy (HIPEC). Proteomic and miRNA measurements used to identify existing drug targets suggested by the genomic/transcriptomic/proteomic characteristics of PMPs, and a particular focus is given on the upregulation of proteins that are known to be drug targets. A stepwise method to select drug candidates as well as appropriate methods for their subsequent mechanistic validation on the in vitro models generated developed.

PIs/Researchers: Dr Jill Barber, Prof Amin Rostami-Hodjegan, Dr Costas Demonacos, Dr Areti-Maria Vasilogianni (post doc)

Funding: CRUK Accelerator Award

The use of PKPD modelling to analyse real-world healthcare data

The project performed the necessary groundwork for pharmacological modelling of RWD. Several identified prerequisites examined: the availability of healthcare datasets in Sweden and the United Kingdom, their variable specifications, data requirements for pharmacological modelling, data management, sharing, and ethics. A workshop held to present the findings and discuss solutions with experts from various domains to produce a feasibility analysis and roadmap for pharmacokinetic/pharmacodynamic (PKPD) analysis of RWD.

PI/researchers: UoM – Dr Kayode Ogungbenro, Prof Leon Aarons; KTH – Adam Darwich

Funding: KTH Royal Institute of Technology and Stockholm University Joint Research Seed Fund