EPSRC Centre for Doctoral Training (CDT) in Molecular Sciences for Medicine (MoSMed)

Small molecule tools to target glucose metabolism in non-alcoholic fatty liver disease

Newcastle University, Chemistry

Supervisory Team

- **Dr Celine Cano (Lead)**
- **Professor Loranne Agius**
- **Professor Ehmke Pohl**

Project overview/context

The project is aligned to our ongoing efforts to synthesise chemical tools to interrogate and modulate glucokinase enzyme (GK). These chemical probes will be used to establish GK inhibition as a therapeutic target in non-alcoholic fatty liver disease (NAFLD). The project will be split into two complimentary strands which will be approached in parallel: a) the design and synthesis of novel proteolysis targeting chimeric molecules (PROTACs) of GK will test the hypothesis that GK can be targeted for degradation; b) the mapping of GK protein by screening the Newcastle FragLites library will assess GK druggability and establish whether GK inhibition is tractable with a small molecule.The studentship will provide a wealth of experience in medicinal chemistry design and organic synthesis along with the opportunity to interact with scientists at AstraZeneca. This would be ideal training for someone wishing to pursue a career in medicinal chemistry or chemical biology.

Research Project

Background

NAFLD is now the leading cause of chronic liver disease and has a 25% prevalence worldwide; yet the overall understanding of the disease remains very limited and no therapeutics are currently available for treatment or prevention. NAFLD covers a range of conditions including hepatic steatosis, and hepatocellular carcinoma. We hypothesise that the raised blood triglycerides could be caused by high hepatic glucose disposal resulting from dietary carbohydrate excess and elevated glucokinase (GK) activity and that GK inhibition in the liver could be an effective therapy for NAFLD.

Aims

We propose to design novel chemical biology tool molecules to the study and manipulation of glucokinase activity. We will test the hypothesis that inhibition of liver glucokinase attenuates hepatic glucose clearance and thereby protects against liver steatosis, raised blood triglycerides and subsequent progression.

Development of Glucokinase PROTACs

A proteolysis targeting chimeric molecule (PROTAC) will provide a proof of concept tool to test the hypothesis that GK can be targeted for degradation. Activators of GK (GKA) have been developed as a potential therapy for type 2 diabetes but no effective drugs have demonstrated sustained glucose lowering. PhD students in the group are currently developing a PROTAC of GK based on established GKA scaffolds. These comprise a ligand for an E3 ligase protein attached by a ‘traditional’ linker (alkyl, PEG, and extended glycol chains) to a ligand for GK. **Our proposal is to expand this work by exploring alternative linker strategies for the development of active PROTAC degraders.**

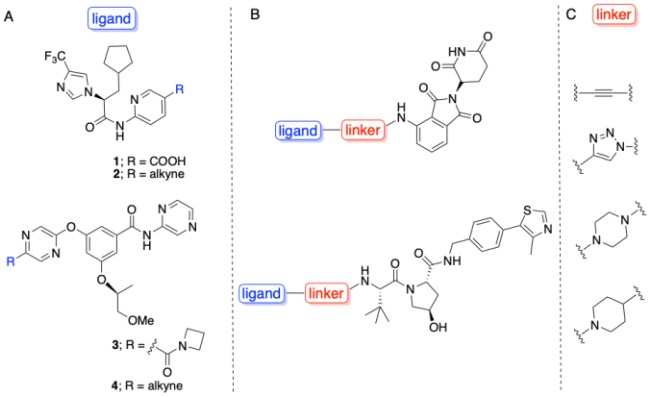

Figure 1: A) Lead GK activators PF-04991532 (1, Pfizer) and AZD1656 (2, AstraZeneca), indicating attachment point(s) for linker (in blue); B) PROTACS conjugated to cereblon (CRBN) and VHL ligands C) novel linkers to be explored.

These novel linkers will provide some rigidity, such as alkynes and heterocyclic scaffolds (e.g., piperazine/piperidines), but will also enable the modulation of the PROTAC physico-chemical properties, in an attempt at reducing the gap between PROTACs and traditional drug-like chemical space (Figure 1). For example, introduction of an ionisable pyridine/di-piperidine motif has the potential to significantly improve aqueous solubility compared to parent PROTACs bearing allhydrocarbon linkers. The triazole moiety also appears as an attractive replacement of traditional linkers. Triazole click chemistry will be used for the combinatorial PROTAC synthesis and rapid identification of anchor-linker-warhead combinations displaying optimal degradation efficiency. For example, we propose to use the copper-catalysed Huisgen 1,3-dipolar cycloaddition reaction to expedite PROTAC synthesis in a highly convergent manner by using an alkyne moiety conjugated to one ligand (for example, GKA derivatives 2 and 4, Figure 1) and an azide conjugated to the other. Photoswitchable PROTACs to incorporate a photolabile “cage” into the PROTAC structure will also be attempted on our system. These various strategies will be explored by the student to obtain diverse libraries of PROTACs with variation in linker length, composition, or conjugation vector.

Synthesised PROTAC tools will be tested in cellular assays already available (Agius’group, NCL, Faculty of Medial Sciences).

Fragments that inhibit liver glucose phosphorylation

The mapping of GK by screening the Newcastle FragLites library will provide an invaluable proof-of-concept study to assess GK druggability and establish whether GK inhibition is tractable with a small molecule. FragLites are a set of halogenated compounds expressing paired hydrogen-bonding motifs that are able to identify productive drug-like interactions within a protein. X-Ray screening of the FragLites library will be conducted in Pohl’s group and the follow-up development of hit compounds directly from the FragLite map will be carried out by the student (Cano’s group). To our knowledge, inhibition of glucokinase with a small molecule has not been reported.

Training & Skills

The student will be based in a dynamic multidisciplinary drug discovery and translational research environment and join a cohort of PhD students working on non-alcoholic fatty liver disease. The Newcastle Medicinal Chemistry Group is a fully integrated drug discovery group, consisting of 30 researchers. The group hosts regular group meetings to discuss progress, as well as medicinal and synthetic chemistry literature reviews. We also hold monthly multidisciplinary project reviews at which the student will be expected to present results to colleagues in Biosciences. Our laboratories house state of the art equipment, including a dedicated 500MHz NMR spectrometer, modern microwave synthesisers, automated chromatography, preparative HPLC and an Agilent 6550 iFunnel QTOF mass spectrometer. Each year, the PhD student will also undertake short (circa 2 months) placements within Bioscience/Pharmacology lab (Agius) and Structural Biology lab (Pohl) to provide further multidisciplinary training.

Further Information

Please contact Lead Supervisor for project enquiries: **celine.cano@ncl.ac.uk**

How to Apply

You must apply through the University’s online postgraduate application system

You will need to:

- Insert the **programme code 8207F** in the programme of study section
- Select ‘**PhD in Molecular Sciences’** as the programme of study
- Input (only) the **studentship reference code (e.g. 21_03)** that you are applying for in the studentship/partnership reference field when prompted (all codes are outlined in the individual project adverts found on the

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MoSMed website: https://research.ncl.ac.uk/mosmed/phdstudentships/)

- Attach all documents that are requested including a CV and cover letter. The cover letter must **clearly** state the project reference code, the full title of the studentship and set out how your interests and experience relate to the project
- Attach degree transcripts and certificates and, if English is not your first language, a copy of your English language qualifications
- Email: mosmed.cdt@ncl.ac.uk once you have submitted your application to confirm the project you have applied for. Please include the studentship reference code and full project title.

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