Proceedings of the British Pharmacological Society at http://www.pA2online.org/abstracts/Vol21Issue1abst004P.pdf

Synthesis and evaluation of novel non-peptide agonists of NPR-C

D. Conole[1], S. H. Myers[1], D. L. Selwood[1], A. J. Hobbs[2], C. Pérez-Ternero[2]. [1]Wolfson Institute for Biomedical Research, University College London, London, United Kingdom, [2]William Harvey Research Institute, Heart Centre, Barts & The London School of Medicine, Queen Mary University of London, London, United Kingdom.

Introduction: Endothelium-derived C-type natriuretic peptide (CNP) possesses cytoprotective and anti-atherogenic functions that regulate vascular homeostasis and smooth-muscle relaxation. The vasoprotective properties of CNP are mediated by the natriuretic peptide receptor type-C (NPR-C), so the latter represents a novel therapeutic target for the treatment of cardiovascular diseases. Thus, we have designed and developed an array of biophysical and physicochemical methods to help design and assess small molecule drug-like mimetics of CNP agonists at NPRC.

Methods: Biophysical techniques such as surface plasmon resonance (SPR), fluorescence polarisation (FP) and thermal shift (TS) assays are employed to assess binding of a distinct structural class of small molecules to NPR-C. Different assay conditions are investigated to explore the feasibility and dynamic range of each method and peptide-based agonists and antagonists are used as controls (e.g. M372049)[2] to validate/invalidate these conditions. In-house kinetic solubility and parallel artificial membrane permeability assays (PAMPA) are also developed and established to identify trends of small molecule drug-like mimetics with respect to their physicochemical profile.

Results: Evaluation of a particular small molecule structural class by SPR showed a range of SAR trends with respect to NPRC binding. Complementary biophysical techniques such as FP and TS showed evidence of natural ligand binding and thus assay validation, but these experiments did not appear sensitive enough to measure non-peptidic small molecule binding. Physicochemical profiling allowed distinction of small molecules more suitable for development as an oral therapy.

Conclusion: We have established new biophysical methods for the assessment of non-peptide small molecule agonists of NPR-C. Some of these methods are high throughput and can be adapted for library screening, whereas others could serve as an orthogonal biophysical technique for conformation of binding. The established in-house physicochemical methods complement the biophysical techniques and thus the overall drug design process. We foresee that such techniques will facilitate the development of potential therapeutic agents for cardiovascular diseases.

References:

1. Moyes, AJ *et al.* (2014). *J. Clin. Investig.* **124**(*9*); 4039-4051.
2. Veale, CA *et al.* (2000). *Bioorg. Med. Chem. Lett.* **10**; 1949-1952