Proceedings of the British Pharmacological Society at http://www.pA2online.org/abstracts/Vol19Issue1abst010P.pdf

Screening of orthosteric antagonists for the pro-inflammatory GPR84 receptor

S. J. Mancini[1], J. Mobarec[2], R. Newman[2], N. Swain[2], M. Barnes[2], A. Tobin[1], G. Milligan[1]. [1]Institute of Molecular, Cell and Systems Biology, University of Glasgow, Glasgow, United Kingdom, [2]Heptares Therapeutics, Welwyn Garden City, United Kingdom.

Introduction: The orphan G protein-coupled receptor GPR84 is reported to be a pro-inflammatory receptor expressed on immune cells and adipocytes, thus there is considerable interest in targeting GPR84 in a range of conditions. The GPR84 antagonist GLPG1205 entered a clinical trial for treatment of ulcerative colitis, but failed due to lack of efficacy[1]. A recent study identified three distinct ligand binding sites within GPR84[2], demonstrating the complex pharmacological regulation of the receptor. The aim of this current study was to identify tool compounds for this currently under-characterised GPCR.

Method: A panel of computationally selected ligands from Heptares Therapeutics, alongside the orthosteric GPR84 agonists embelin and compound-1 (2-(hexylthiol) pyridimine-4,6 diol)[3] were screened against GPR84. Membranes were prepared from Flp-In™ T-REx 293™ cells stably expressing a doxycycline-inducible GPR84-G_{i2} C352I fusion protein and utilised in a [^{35}S]GTPγS incorporation assay to assess receptor activation and mechanism of action. GPR84 was computationally modelled and small-molecules were docked and simulated utilizing the Schrödinger suite of computational chemistry programs.

Results: Of a panel of >500 selected ligands, fifteen were found to antagonise agonist-stimulated GPR84 activation >75%. Three of these compounds had IC50 values in the micromolar range (pIC50 = 5.3, 5.4, 5.6) and were selected for further characterisation. Mechanism of action studies demonstrated that two of these compounds were competitive with the orthosteric agonist compound-1, while all were non-competitive with the allosteric agonist PSB-16671 (3,3'-di-(5,7-difluoro-1H-indol-3yl)methane)[4]. GPR84 was modelled bound to agonists and selected antagonists, providing a molecular framework for the mode of action of these ligands which will be experimentally validated.

Conclusion: Screening of computationally selected ligands at GPR84 revealed two novel surrogate ligands which function as orthosteric antagonists at GPR84. These tool compounds may be utilised to further explore the biology and therapeutic potential of this hard-to-target receptor.

References:

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