RCoA Research, Education & Travel Grants 2013

Award: The Ernest Leach Fund

Applicant: Dr Mhamad Ghiyath Al-Hashimi

Project Title: Investigating immune-modulatory actions of Nociceptin/Orphanin FQ (N/OFQ) system in peripheral human blood

Project Description

There is a body of evidence supporting that clinical and recreational use of opioids can modulate the innate and adaptive immune function with some degree of controversy regarding the exact mechanisms of such effect.[1] Opioid receptors are classified as MOP (μ, mu), DOP (δ, delta) and KOP (κ, kappa) or classical naloxone sensitive receptors and NOP (receptor for nociceptin/orphanin FQ [N/OFQ]) which is naloxone insensitive.[2] Opioids in current medical practice act mainly as agonists on classical opioid receptors (MOP, KOP, DOP).[1] It has been suggested that opioid induced immune modulation may occur via direct action on immune cells themselves. Several previous studies suggested the expression of classical opioid receptors on peripheral blood mononuclear cells (PBMC's)[1,3]; However the presence of opioid receptors in peripheral individual immune cells was evaluated by our group in series of experiments [4-6] failing to detect MOP, KOP, and DOP receptors but identified NOP receptor on immune cells and peripheral whole human blood. PCR NOP mRNA transcripts detected by our group were strongly expressed and consistent leading to the conclusion that NOP functioning receptor protein is present in peripheral human blood.

There is a relative lack of functional information and the significance of NOP receptor on immune cells, their involvement in immune modulation/regulation, and whether or not the N/OFQ-NOP system can be a potential future immune modulatory therapeutic route.

We intend to conduct series of experiments to study the functional effect of NOP activation on peripheral immune cells (native from volunteers and cell line based). Cells will be incubated with N/OFQ (endogenous peptide agonist), UFP-112 (synthetic peptide agonist), Ro64-6198 (non-peptide agonist), UFP-101 (peptide antagonist/partial agonist) SB612111 (non-peptide antagonist) at clinically relevant concentrations (picomolar –nanomolar range) and in various combinations. We will determine efficacy and functional potency for both agonists and antagonists. Our read-out will be cell migration and this will be assessed using the transwell assay format. We will also quantify various cytokines (including GM-CSF, IFN-γ, IL-1β, IL- 2, IL-(4-6), IL-(8-10), and TNF-α) to link cell migration/activation with cytokine production. This work will form part of my MD project with the University of Leicester.

We will use grant funds to cover the cost of migration assay, consumables, and human cytokine quantification kits (as detailed below). We have the infrastructure and personnel to finish this research project within 3 months of funds availability. We intend to submit the results for publication in peer reviewed medical journals and present the results at national meetings. In addition this functional data will be used as part of an application for more substantial funding as an MRC project grant.

References

1- Al-Hashimi M, Scott SW, Thompson JP, Lambert DG. Opioids and immune modulation: more questions than answers. *Br J Anaesthesia* 2013; **111**(1):80-8

2- Lambert DG. The Nociceptin

3- Cedat P, Mantione K, Bilfinger TV, Stefano GB. Real-time PT-PCR measurement of the modulation of Mu opioid receptor expression by nitric oxide in human mononuclear cells. *J Immunol* 2001; **170**: 1123-8
4- Williams JP, Thompson JP, McDonald J, Barnes TA, Cote T, Rowbotham DJ, Lambert DG. Human peripheral blood mononuclear cells express nociception/orphanin FQ, but not mu, delta, or kappa opioid receptors. *Anaesthesia Analgesia* 2007; **105**(4): 998-1005
5- Williams JP. Opioids and a Neuro-Vascular-Immune axis. University of Leicester doctoral thesis 2008. http://hdl.handle.net /2381/4160 (last accessed 4/Feb/2013)
6- M. Al-Hashimi, J. McDonald, J.P. Thompson and D.G. Lambert. Classical opioid