Proceedings of the British Pharmacological Society at http://www.pA2online.org/abstracts/Vol18Issue1abst187P.pdf

Evidence for mitochondrial involvement in Ipratropium bromide mediated myocardial injury

F. A. Khalefah, A. Hussain, B. Burke, K. L. Harvey. School of Life Sciences, Coventry University, Coventry, United Kingdom.

Introduction: Ipratropium bromide (IP), a non-selective muscarinic receptor antagonist, is administered in treatment regimens for Chronic Obstructive Pulmonary Disease (COPD). It has been indicated, clinically, that COPD patients with underlying Ischaemic Heart Disease (IHD) develop higher levels of systemic pathologies, specifically adverse cardiovascular events (CVEs) (1,2). Previous work in our laboratory has shown that IP increases myocardial injury via both apoptosis and necrosis in clinically relevant *in vitro* models of ischaemia/reperfusion (I/R) (3). However, despite evidence to suggest this has mitochondrial involvement, the mechanism of this drug-induced toxicity is yet to be fully elucidated. The aim of the current study is to investigate whether the cytotoxic effects of IP involves mitochondrial fission and fusion via assessment of IP induced myocardial damage using a Langendorff model of I/R subjected to co-administration of IP with Mdivi-1 (Mitochondrial division inhibitor).

Methods: Adult, male, Sprague-Dawley rats were sacrificed via cervical dislocation and hearts were immediately placed onto Langendorff perfusion apparatus. Following 20 minutes stabilisation, hearts were subjected to 35 minutes left ventricular ischaemia via ligation of the descending left coronary arteries. Reperfusion was initiated and lasted 120 minutes. IP (1, 10 and 100 nM) ± Mdivi-1 (100 nM) was administered at the start, and throughout, reperfusion. Infarct sizes were assessed via Evans Blue staining and 2,3,5-triphenyl tetrazolium chloride exclusion treatment to delineate between live and infarcted tissue within the ischaemic area.

Results: Results have indicated that IP significantly increases infarct size ($p < 0.05$ for all concentrations vs. control) thus supporting previous literature. The administration of 100 nM Mdivi-1 showed cardio-protection via a reduction in infarct size compared with the untreated control (29.1 ± 4% vs. 44.3 ± 1% (control), $p< 0.001$). When IP (100 nM) was co-administered with Mdivi-1 (100 nM) importantly this demonstrated a significant reduction in infarction compared with the untreated control (36.8 ±3% vs. 44.3 ± 1% (control), $p <0.05$), thus abrogating the IP mediated increase in myocardial injury in this model. n= 6 for all experiments.

Conclusions: This study supports previous findings that IP exacerbates myocardial injury, but is the first study to more specifically relate a mitochondrial pathway to IP induced myocardial injury.

References:

1. Ogale, S. S., et al (2010) *Chest* **137(1):** 13-9
2. Finegold, J. A., et al (2013) *Int J Cardiol.* **30;168(2):**934-45
3. Harvey, K. L., et al (2014) *Toxicol Sci.* **138(2):**457-67