Abstract 178 Table 2

Cardiac Morphology and Fibrosis	Control	ISO		
		Immunocompetent	Immunocompromised	P value
Fibrotic Mass (mg)	0	16±1	35±5	0.02

Results ISO caused changes in: 1) cardiac function – increasing end diastolic and end systolic volumes while decreasing ejection fraction, 2) cardiac morphology – increasing the left ventricular mass resulting in hypertrophy (enlargement of the heart), and 3) cardiac fibrosis – apex of the heart being the most vulnerable. A differential-distribution of fibrosis-severity across the heart has also been detected.

Discussion In agreement with initial hypothesis, there was a differed-response between Immunocompetent and Immunocompromised subgroups: the Immunocompetent showed a trend towards cardiac hypertrophy, whereas the Immunocompromised were more vulnerable to fibrosis. This cardioprotective role of the immune system could be due to CXCR4 signalling that is also implicated in cancer-metastasis, stem-cell-migration, angiogenesis, and haematopoiesis, which we will follow up.

Conclusion ISO-murine-disease-model has been established and characterised to study myocardial damage, with cardiac function, morphology, and damage in the form of fibrosis detected, visualised, and quantitatively assessed using cardiac MRI. ISO caused changes in cardiac function, morphology, and fibrosis. There was also a differential-distribution of fibrosis-severity across the heart as shown visually by high-resolution sequential MRI images.

179 PHOSPHODIESTERASE-5 INHIBITION WITH SILDENAFIL SUPPRESSES CALCIUM WAVES BY REDUCING SARCOPLASMIC RETICULUM CONTENT

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Rationale: Occurrence of diastolic Ca^{2+} waves in cardiac myocytes leads to arrhythmias by inducing delayed after-depolarisations. Waves are initiated when sarcoplasmic reticulum (SR) content reaches a critical threshold level. The phosphodiesterase-5 inhibitor sildenafil (Sil) is antiarrhythmic in mammalian myocardial ischaemia models, while Sil reduces Ca^{2+} transient amplitude and sarcoplasmic reticulum (SR) Ca^{2+} content in rat myocytes.

Objective: To determine effects of Sil on propensity to Ca^{2+} waves in the large mammal.

Methods: Sheep ventricular myocytes were voltage clamped and intracellular Ca^{2+} measured using Fura-2. Cells were paced at 0.5 Hz with depolarisations from −40 mV to +10 mV. When at steady state, waves were induced with 10–15 mM Ca^{2+}. Upon regular waving, Sil (1μM) was applied. To determine threshold SR content, caffeine (10 mM) was added immediately following a wave, and both wave and caffeine-induced I_{NCX} integrated. Differences between groups were determined using students paired t tests.

Results: Increasing external Ca^{2+} to 10–15 mM increased SR content and induced diastolic waves. Sildenafil abolished waves in 9/15 cells. In cells where Sil terminated waves, SR content was reduced below threshold. In addition, Sil treatment was associated with a reduced rate constant of SERCA (k_{SERCA}−66.0±9.9% of control, $p<0.005$), an initial (first 4 s) increase in sarcolemmal efflux via the I_{NCX} tail current (+142 ± 36.4%, $p<0.01$), and reduced sarcolemmal influx via $I_{Ca\text{-}L}$ (–30.5±5.6%, $p<0.005$). In cells continuing to wave in Sil, SR threshold for waves was unchanged (126.9 $\mu molL^{-1}$ ctrl vs 147.2 $\mu molL^{-1}$ Sil, $p=0.6$). In unstimulated cells spontaneously waving in 10–15 mM Ca^{2+}, sildenafil reduced wave frequency (6.3 waves per 20 s vs 2.7, $p<0.005$). The protective effect of sildenafil on both wave models was abolished when cells were pre-incubated with the PKG inhibitor, KT5823. Sildenafil suppression of waves was also observed in cells from animals in end-stage heart failure, while Sil suppressed ventricular ectopy and episodes of torsades de pointes *in vivo* in a sheep model of LQT2.

Conclusions: Sildenafil suppresses waves induced by elevated external Ca^{2+} via a PKG-dependent mechanism, and mediated by a reduction in SR content, which itself is caused by reduced SERCA function ± reduced $I_{Ca\text{-}L}$. These findings highlight novel antiarrhythmic properties of PDE5 inhibition and translate to suppression of triggered arrhythmias *in vivo*.

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180 THE AORTA CAN ACT AS SITE OF T CELL PRIMING AND PROMOTES A LOCAL CD4+ ADAPTIVE IMMUNE RESPONSE IN EARLY STAGE ATHEROSCLEROSIS IN APOLIPOPROTEIN-E-/- MICE

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Artery tertiary lymphoid organs (ATLOs) in the adventitia adjacent to intimal plaque in aortas from aged apolipoprotein-E (apoE)$^{-/-}$ mice have recently been shown by us to orchestrate the aortic immune response in the advanced stages of experimental atherosclerosis, highlighting the importance of the vascular immune response and its contribution to the pathology. Antigen presenting cells (APCs) and T cells are found in both human and animal naÃƒÂ¯ve and atherosclerotic vessels; however, the mechanisms leading to T cell activation in the arterial wall remain poorly understood. Here we utilised flow cytometry to present a quantitative assessment of the major antigen presenting cells in both C57BL/6 wild type (WT) and apoE$^{-/-}$ mice in the aorta, aortic draining lymph nodes and spleen. By employing a model antigen and antibody detections system (Eα-GFP/Y-Ae), it was possible to assess the ability of aortic APC subsets to present antigen *in vivo*. We also performed a comprehensive phenotypic analysis of CD4^{+} T cells in healthy versus atherosclerotic aorta using surface marker expression and cytokine signatures. This study revealed that aortas from atherosclerotic mice contained more CD4^{+} T cells and more Th1 T cells (IFN-γ^{3}+) compared to WT, as well as showing a phenotypic switch from a nave to an activated phenotype. On the contrary, changes in T cell phenotype in the spleen and draining lymph nodes between WT and apoE$^{-/-}$mice were either equivalent or modest. One hypothesis regarding the presence of T cell autoreactive clones