morphological abnormalities in E15.5 hearts. Future work will identify how altered protein functions cause these changes.

BS31 MARK4 CONTROLS ISCHAEMIC HEART FAILURE THROUGH MICROTUBULE DETYROSINATION

Xuan Li. *University of Cambridge, Level 5, Box 157 Addenbrooke's Hospital, Cambridge, CAM CB2 0QQ, UK*

10.1136/heartjnl-2022-BCS.211

Myocardial infarction (MI) is a major cause of premature adult death. Compromised cardiac function after MI leads to chronic heart failure with systemic health complications and high mortality rate. Effective therapeutic strategies are highly needed to improve the recovery of cardiac function after MI. More specifically, there is a major unmet need for a new class of drugs that improve cardiomyocyte contractility, because currently available inotropic therapies have been associated with high morbidity and mortality in patients with systolic heart failure, or have shown a very modest risk reduction. Microtubule detyrosination is emerging as an important mechanism of regulation of cardiomyocyte contractility. By using combined approaches (genetics, animal disease model, physiology, advanced super-resolution microscopy, biochemical fractionation and etc), we show that deficiency of Microtubule-Affinity Regulating Kinase 4 (MARK4) substantially limits the reduction of left ventricular ejection fraction (LVEF) after acute MI in mice, without affecting infarct size or cardiac remodeling. Mechanistically, we provide evidence that MARK4 regulates cardiomyocyte contractility through promoting microtubule-associated protein 4 (MAP4) phosphorylation, thereby facilitating the access of Vasohibin 2 (VASH2), a tubulin carboxypeptidase (TCP), to microtubules for alpha-tubulin detyrosination. Our results show how cardiomyocyte microtubule detyrosination is finely tuned by MARK4 to regulate cardiac inotropy, and identify MARK4 as a promising druggable therapeutic target for improving cardiac function after MI.

BS32 INTEROGATING THE INTERPLAY BETWEEN MATRIX TOPLOGY, MATRIX STIFFNESS AND AORTIC SMOOTH MUSCLE CELL FUNCTION

[1]Teclino Afewerki, [2]Robert Johnson, [3]Reesha Solanki, [4]James Courtney, [3]Derek Warren. *[1]School of Pharmacy, University of East Anglia, School of Pharmacy Norwich Research Park Norwich, NFK NR4 7TJ UK; [2]Universit of East Anglia; [3]School of Pharmacy, University of East Anglia; [4]School of Chemical, University of Birmingham*

10.1136/heartjnl-2022-BCS.212

Background Vascular smooth muscle cells (VSMCs) are the predominant cell type in the arterial wall and normally adopt a quiescent, contractile phenotype to regulate vascular tone. VSMCs are exposed to multiple mechanical cues, including stretch and matrix stiffness, which regulate VSMC contraction. Recent studies have shown that extracellular matrix (ECM) topology and stiffness influences migration of a variety of cell types. Whilst we have extensive knowledge of how soluble factors regulate VSMC function, our understanding of the importance of matrix-derived cues is limited. Methodes In this study we use smooth and grooved polyacrylamide hydrogels of physiological and pathological stiffness, to investigate the interplay between matrix topology, matrix stiffness and VSMC function.

Results VSMCs grown on grooved hydrogels of physiological stiffness were less spread than those grown on smooth hydrogels. Traction force microscopy revealed that VSMCs on the grooved hydrogels of physiological stiffness generated enhanced traction stress compared to their counterparts on smooth hydrogels. VSMCs on grooved hydrogels of pathological stiffness still generated enhaved traction stress however, they displayed similar spreading to VSMCs grown on smooth hydrogels. Finally, we tested the impact on migration. VSMCs on grooved hydrogels of physiological stiffness displayed a reduced migrational capacity compared to their conuterparts on smooth hydrogels. However, VSMC migrational capacity remained unaltered between grooved and smooth hydrogels of pathological stiffness. Conclusion These data demonstrates that matrix topology differentially regulates VSMC function at physiological and pathological stiffness due to increased contraction of their surrounding environment. Through reducing VSMC force generation, we can potentially delay to onset of a range of age-related cardiovascular diseases.

BS33 CYCLIC-AMP INCREASES NUCLEAR ACTIN MONOMER WHICH PROMOTES PROTEASOMAL DEGRADATION OF RELA/P65 LEADING TO ANTI-INFLAMMATORY EFFECTS

[1]Mark Bond, [2]Joseph Hawkins, [2]Madeleine McNeill, [2]Reza Ebrahimighaei, [2]Harry Mellor, [2]Andrew Newby. *[1]University of Bristol, Translational Health Sciences, University of Bristol, Bristol, BST BS2 8HW, UK; [2]University of Bristol*

10.1136/heartjnl-2022-BCS.213

The second messenger, 3', 5'-cyclic adenosine monophosphate (cAMP) has potent anti-inflammatory actions. These have been attributed to the ability of cAMP-induced signals to interfere with the function of the proinflammatory transcription factor Nuclear Factor-kappa B (NF-κB). However, the mechanisms underlying the modulation of NF-kB activity by cAMP remain unclear. Here we demonstrate an important role for cAMP-mediated increase in nuclear actin monomer levels in inhibiting NF-κB activity. Elevated cAMP or adenovirus mediated expression of a nuclear localised polymerisation defective actin mutant (NLS-ActinR62D) inhibited basal and TNFα induced mRNA levels of NF-κB-dependent genes and NF-κB-dependent reporter gene activity. Elevated cAMP or NLS-ActinR62D did not affect NF-κB nuclear translocation but did reduce total cellular and nuclear RelA/p65 levels. Preventing the cAMP induced increase in nuclear actin monomer, either by expressing a nuclear localised active mutant of the actin polymerising protein mDIA, silencing components of the nuclear actin import complex IPO9 and CFL1 or over expressing the exportin XPO6, rescued RelA/p65 levels and NF-κB reporter gene activity in forskolin stimulated cells. Elevated cAMP or NLS-ActinR62D reduced the half-life of RelA/p65, which was reversed by the proteasome inhibitor MG132. Accordingly, forskolin stimulated association of RelA/p65 with ubiquitin affinity beads, indicating increased ubiquitination of RelA/65 or associated proteins. Taken together, our data demonstrates a novel mechanism underlying the anti-inflammatory effects of cAMP and highlights the important role played by nuclear actin in the regulation of inflammation.