Novel endothelial-enriched long non-coding RNA AL590004.3 (IRENE) is dysregulated in HFpEF and regulates endothelial enhancement of cardiomyocyte function

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Funding Acknowledgements: Type of funding sources: Public grant(s) – EU funding. Main funding source(s): ERC consolidator

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Background: Heart failure with preserved ejection fraction (HFpEF) is the most prevalent form of HF to date. However, there is a lack of medication which stages HFpEF as an unmet clinical need. HFpEF is characterized by cardiac microvascular dysfunction, promoted by co-morbidities-associated low-grade pro-inflammatory state. We have shown that cardiac endothelial cells regulate cardiomyocyte relaxation, which is impaired when endothelial cells are inflamed. Long non-coding RNAs (lncRNAs) have been implicated in various cardiovascular diseases. However, their role in HFpEF is unknown. Here, we discovered a novel lncRNA AL590004.3 which is significantly reduced in the heart of patients with HFpEF. We hypothesized that AL590004.3 is important for cardiac endothelial function, and dysregulation of endothelial AL590004.3 impairs cardiomyocyte relaxation.

Methods and results: We demonstrated that AL590004.3 is significantly downregulated (~7 fold, p<0.001) in HFpEF as assessed by in silico analysis of RNA sequencing of HFpEF patient cardiac biopsies. To assess the tissue distribution, we performed RNA sequencing comparing cells from various tissues and found the highest expression of AL590004.3 in the cardiac microvascular endothelial cells (CMECs), indicating its importance in cardiac endothelial cell biology. The expression of AL590004.3 was reduced when CMECs were exposed to pro-inflammatory cytokine TNFα and IL1β, indicating the detrimental effect of inflammation, as displayed by patients with HFpEF. Further, replicative senescent of CMECs led to reduced AL590004.3 expression (2 fold, p<0.005), showing the deleterious contribution of ageing, similar to what occurs in HFpEF patients. We showed that AL590004.3 is mostly expressed in the chromatin fraction (~90%) of CMECs, suggesting its function in chromatin modification. To assess the function of AL590004.3, we performed loss-of-function. Using endothelial-cardiomyocyte co-culture set-up, we showed silencing of AL590004.3 in CMECs impaired endothelial function to enhance cardiomyocyte relaxation (~1.5 fold, p<0.01).

To dissect the molecular mechanism, we performed RNA sequencing of CMECs devoid of AL590004.3 and revealed four protein-coding genes (PXDC1, FAM50, ECI2, PRPF4B) that are differentially co-regulated, indicating their potential as protein-binding partners, which warrant further investigation.

Conclusion: We showed that AL590004.3 is a cardiac endothelial cell-specific lncRNA that is significantly downregulated in cardiac biopsy from HFpEF patients. AL590004.3 is important for cardiac endothelial cell function as loss-of-function in CMECs impaired endothelial enhancement of cardiomyocyte relaxation. Herewith, we name this lncRNA Important for the Regulation of ENDothelial function in HFpEF (IRENE).