An Experimental and Computational Analysis of Selective Class I HDAC Inhibition in Pathological Cardiomyocyte Hypertrophy

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Abstract
Pathological cardiomyocyte hypertrophy is a major contributor to heart failure that is regulated by histone deacetylases (HDACs), which exert both pathological and protective roles. Therefore, this study investigates whether selectively inhibiting Class I HDACs provides a more effective and targeted approach towards preventing pathological cardiomyocyte hypertrophy as compared to non-selective inhibition. To address this, human induced pluripotent stem cell-derived cardiomyocytes will be used to model hypertrophy, with outcomes assessed through cell size, gene expression, and functional assays. In addition, a computational model will be used to simulate HDAC-regulated signalling pathways and support the evaluation of experimental findings.

Introduction

Pathological cardiomyocyte hypertrophy, one of the most common genetic heart conditions, is a major contributor to heart failure worldwide. Estimated to affect approximately 1 in 500 individuals (0.2% of the world), with many cases remaining undiagnosed, it is of major importance that methods of treating this issue are determined (Semsarian et al., 2015). At its core, this disease results from changes in gene expression within heart cells. These changes are controlled by histone deacetylases (HDACs) that modify chromatin structure thereby influencing gene expression (Haberland et al., 2009). In the heart, HDACs take on opposing functions, where Class I HDACs promote hypertrophic growth, while Class IIa HDACs help protect against this, by binding to and inhibiting MEF2, preventing the activation of genes that drive pathological cardiomyocyte growth (McKinsey et al., 2002; Lu et al., 2000). Although HDAC inhibition has been explored as a potential therapeutic strategy, most current inhibitors are non-selective and target multiple HDAC classes, both beneficial and harmful, simultaneously. This reduces efficacy and increases toxicity (Yao et al., 2021). Therefore this study aims to selectively inhibit Class I HDACs, while preserving Class IIa HDAC function, using both experimental and computational methods, to provide a more effective approach to reduce pathological cardiomyocyte hypertrophy.

Literature Review

Current research has highlighted the central role of HDACs in pathological cardiomyocyte hypertrophy; however, it remains limited by a reliance on non-selective inhibition strategies and insufficient integration of computational modelling approaches (Yao et al., 2021; Haberland et al., 2009). HDACs play a key role in regulating cardiac gene expression and remodelling processes that contribute to hypertrophy (Haberland et al., 2009). A recent article demonstrates the extensive therapeutic potential of HDAC inhibition in cardiac disease models, by virtue of its ability to mitigate hypertrophy and regulate pathological gene expression, thereby leading to an overall improvement in cardiac function (Lu et al., 2024). However, these findings remain inconsistent due to the use of non-selective inhibition strategies, which can suppress both pathological and protective HDAC functions, leading to uncertainty regarding long-term safety (Lu et al., 2024; McKinsey et al., 2002). Furthermore, HDAC inhibitors have been shown to cause adverse cardiac effects including arrhythmias, reduced heart rate, and contractile dysfunction, with pan-HDAC (non-selective) inhibitors producing stronger effects compared to more selective inhibitors (Kopljar et al., 2016). This highlights a key limitation in current research: a failure to account for the functionally opposing roles of HDAC classes. These approaches lack specificity, limiting their clinical applicability. This emphasises the need for computational modelling approaches.

Methodology

Human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) will be cultured under standard conditions, replicating key aspects of the human physiological environment. Cells will be divided into four groups: a control group (no hypertrophic stimulation), a hypertrophy group (hypertrophic stimulation only), a selective Class I HDAC inhibitor group (hypertrophic stimulation with Class I HDAC inhibitors), and a pan-HDAC inhibitor group (hypertrophic stimulation with non-selective HDAC inhibitors). Pathological hypertrophy will be induced in the appropriate groups using angiotensin II, an established agent that stimulates stress-related cardiac signalling pathways (Kang et al., 2015)

Cardiomyocyte hypertrophy will be quantified using fluorescence microscopy and image-based measurement of cell surface area, a key indicator of pathological remodelling. Gene expression will be assessed using quantitative PCR (qPCR) for hypertrophic markers including ANP, BNP, and β-myosin heavy chain (β-MHC), which are well-established indicators of cardiac stress (Haberland et al., 2009). HDAC regulated pathways, such as MEF2 signalling, will also be evaluated. As disrupted calcium regulation is a defining feature of hypertrophic cardiomyocytes, calcium handling assays will be conducted to assess contractile function. Furthermore, cell viability assays will be performed to evaluate potential cytotoxic effects of HDAC inhibition. These assays measure the proportion of metabolically active, living cells following treatment, providing an indication of overall cell health. This ensures that any observed reductions in hypertrophy or changes in gene expression can be attributed to the specific effects of HDAC inhibition rather than decreased cell viability.

A computational model will be developed to simulate HDAC-regulated signalling pathways involved in pathological cardiomyocyte hypertrophy. The model will represent key molecular interactions between Class I HDACs, Class IIa HDACs, the transcription factor MEF2, and hypertrophic genes including ANP, BNP, and β-MHC, based on established HDAC–MEF2 regulatory mechanisms (Backs et al., 2006; Zhang et al., 2002). These biological relationships will be translated into mathematical rules that describe how changes in HDAC activity influence MEF2 signalling and subsequent gene expression. Alongside this simulations will be conducted under conditions corresponding to the experimental groups used previously, generating predicted outputs such as gene expression levels and relative hypertrophic response over time. This computational implementation will be carried out using Python, supported by pathway databases such as KEGG. Experimental data obtained from gene expression analysis, cell size measurements, and functional assays will be used to calibrate and validate the model. Moreover, agreement between predicted and observed outcomes will be assessed to evaluate model accuracy and refine underlying pathway assumptions, consistent with systems biology modelling approaches in cardiac research (Saucerman et al., 2019; Yang & Saucerman, 2012).

Conclusion

In conclusion, this study aims to determine whether selectively targeting Class I histone deacetylases provides a more effective and precise strategy for reducing pathological cardiomyocyte hypertrophy than non-selective inhibition. By combining controlled in vitro experiments with computational modelling of HDAC-regulated signalling pathways, the research is expected to show that selective inhibition more effectively suppresses hypertrophic gene expression while preserving cardiomyocyte function and viability. Overall, this approach may offer clearer insight into the distinct roles of HDAC classes and support the development of more targeted therapies for cardiac disease.

References

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