Beyond clinical variables: Machine learning integration of clinical and contextual factors for predicting heart failure readmissions

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Abstract

Heart failure patients experience frequent 30-day readmissions, and existing clinical models predict these moderately well. This study used machine learning to integrate clinical and socioeconomic factors to improve prediction accuracy. De-identified electronic medical records from the University of Missouri–Columbia Snowflake system (up to 2025) were used to identify 33,423 adult heart failure patients and 66,804 hospital admissions. Baseline demographic, socioeconomic, clinical, laboratory, medication, comorbidity, and mortality data were extracted. The cohort was split at the patient level into training (70%, n = 46,708), test (20%, n = 6,685), and validation (10%, n = 3,343) sets. After preprocessing, logistic regression, random forest (RF), and XGBoost models were developed. Performance on the test set was evaluated using area under the receiver operating characteristic curve (AUROC), precision–recall AUC (PR-AUC), Brier score, log loss, accuracy, precision, recall, F1 score, specificity, and negative predictive value (NPV), all with 95% confidence intervals (CIs). On the test set, random forest and XGBoost outperformed logistic regression, achieving higher discrimination (AUROC 0.705, 95% CI: 0.688–0.720 and 0.704, 95% CI: 0.688–0.719, respectively) than logistic regression (AUROC 0.684, 95% CI: 0.667–0.701), with improvements in PR-AUC (random forest 0.399, 95% CI: 0.361–0.436; XGBoost 0.394, 95% CI: 0.357–0.426; logistic regression 0.369, 95% CI: 0.332–0.402). Random forest showed superior calibration with the lowest Brier score (0.153, 95% CI: 0.146–0.159) and log loss (0.475, 95% CI: 0.460–0.489).

XGBoost achieved the highest accuracy (0.622, 95% CI: 0.612–0.632) and specificity (0.607, 95% CI: 0.592–0.620), while random forest achieved the highest recall (0.760, 95% CI: 0.737–0.782). Overall, preliminary ensemble machine learning models achieved improved prediction of 30-day readmissions (AUROC ≈ 0.70; RF: Brier ≈ 0.15), supporting use for clinical risk stratification. Further feature engineering and model refinement are planned.

Advancing cardiovascular research through rat models: The rat resource and research center

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Abstract

Rats are often the preferred model organism in cardiovascular research because of their genetic and physiological similarities to humans, their ideal size for surgeries and repeated measurements and the availability of genetically modified strains/stocks that model human conditions and diseases. The NIH-funded Rat Resource and Research Center (RRRC) is a centralized repository for maintaining, archiving and distributing rat models and providing rat-related services to the biomedical community. Currently, the RRRC has over 625 rat lines, including more than 100 with cardiovascular phenotypes such as hypertension. Our website (www.rrrc.us) allows user-friendly navigation to identify models of interest. The RRRC provides expertise in rat colony management, health monitoring, genetic assay development/optimization, and can collaborate on investigator projects and grants. As experts in comparative medicine, with access to skilled laboratory animal veterinarians, we can assist with surgical techniques and perform all aspects of in

vivo rat studies. Fee-for-service capabilities include a wide variety of genetic analyses, strain rederivation and cryopreservation, isolation of rat tissues, custom breeding/colony management, microbiota analysis and characterization of rat models. The RRRC can make genetically engineered rats from start to finish using state-of-the-art technologies such as genome editing and traditional methods like random transgenesis and embryonic stem cell microinjection. New initiatives include supplying induced pluripotent stem cells (iPSCs) to support new approach methodologies (NAMs). The RRRC provides the in vivo models needed to validate NAMs and support studies that require complex biological systems for appropriate scientific rigor and reproducibility. The University of Missouri is home to the NIH-funded MU Rat Testing Center for Somatic Cell Genome Editing, MU Mutant Mouse Resource and Research Center (MMRRC), National Swine Resource and Research Center (NSRRC), MU Animal Modeling Core, and MU Metagenomics Center. Together, these collaborative groups offer diverse animal model-related services across species to advance biomedical and translational research.

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Transient developmental dysregulation of microglial and synaptic pruning markers in the nucleus accumbens of selectively bred low voluntary running rats

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Background

Microglial maturation and their interactions with neurons serve crucial roles during developmental stages, such as synaptogenesis, synaptic pruning, and maintaining brain homeostasis. Evidence suggests that interference with microglial function in the nucleus accumbens (NAc) during adolescence disrupts synaptic pruning and influences adult social behavior in a sex-specific manner. Our previous studies showed that selectively bred low voluntary running (LVR) rats had decreased spine density and numbers of medium spiny neurons (MSN) in the NAc during adolescence, which may underlie altered synaptic function in the motivation for physical activity. However, whether microglia-neuron interactions affect the motivation for voluntary physical activity remains poorly understood. The aim of this study is to determine the developmental profile of microglial and synaptic pruning markers in the NAc of LVR rats.

Methods

We used wildtype Wistar and LVR rats (generations 27-29) in this study. The NAc was collected at postnatal days 0 (P0), 14 (P14), 21 (P21), 35 (P35), and 60 (P60) (N=6-8/sex/timepoint from 2-3 families). We performed quantitative reverse-transcription PCR to examine the RNA expression levels of microglia-specific, dopamine receptor, and synaptic pruning-related genes.

Results

Our preliminary data showed that male LVR rats had significantly increased mRNA expression of microglial reactivity (Iba1), neuron-microglial signaling (Cx3cl1, Cx3cr1), complement regulation (Csmd1), and dopamine receptors (Drd1, Drd2) at P21 and P35 but not P60. There were no differences in the markers of microglial homeostasis (Tmem119), astrocyte activation (Gfap), or classical complement factors (C1qb, C4A).

Conclusion

Our preliminary findings indicate a transient, non-inflammatory dysregulation of the neuroimmune axis during the critical adolescence window, coinciding with our previously reported MSN loss in LVR rats. This altered microglia-neuron interaction during development may play a pivotal role in impairing voluntary physical activity motivation.

Expression of the ERG transcription factor in Doxorubicin-treated endothelium

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Abstract

Doxorubicin (DOX) is an anthracycline chemotherapeutic whose clinical utility is limited by dose dependent cardiotoxicity. Emerging evidence suggests endothelial dysfunction contributes to DOX associated vascular injury. The ETS related gene (ERG) is an endothelial transcription factor required for vascular integrity and homeostasis and is regulated by transforming growth factor β (TGF-β) signaling. We investigated whether DOX alters ERG expression in endothelial cells and whether inhibition of TGF-β signaling modulates ERG regulation during drug exposure and post-treatment recovery.

Human umbilical vein endothelial cells (HUVECs) were treated with DOX (16nM), the TGF-β type I receptor inhibitor SB431542 (1μM), DOX plus SB, or control. Treatments were applied either concurrently for 48 hours or followed by a washout period with SB or vehicle to assess recovery following DOX exposure. ERG protein expression was assessed by quantitative PCR and immunoblotting. In vivo, ERG expression was evaluated by immunohistochemical staining of cardiac tissue from male and female C57BL/6 mice treated with saline, DOX, SB, or DOX plus SB.

Under concurrent treatment conditions, DOX with or without SB did not significantly alter ERG expression in HUVECs compared to controls. In contrast, DOX alone decreased ERG expression during the washout period, whereas SB administration during post-DOX washout was associated with partial restoration of ERG expression relative to DOX-only conditions. Analysis of cardiac tissue from both male and female mice revealed no significant differences in ERG staining intensity between treatment groups. Cardiac tissue was harvested three weeks after final DOX injection, which may allow recovery of ERG abundance in endothelial cells.

These findings indicate that ERG regulation following DOX exposure is context and timing-dependent, with recovery phase modulation of TGF-β signaling influencing endothelial ERG expression. Lack of detectable changes in cardiac ERG after completion of Dox therapy may indicate transient suppression followed by recovery of ERG expression.

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Role of alpha1 adrenergic signaling in nucleus ambiguus regulation of heart rate

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Abstract

The brainstem's nucleus ambiguus (NA) is a central regulatory region for heart rate (HR) through cardiac vagal motor neurons (CVNs) that send direct axonal projections to cardiac tissue. Given their quiescent nature, receptor signaling in CVNs is critical for HR regulation and this regulation is dampened in cardiovascular disease (CVD). Adrenergic receptors (ARs) initiate diverse intracellular signaling based on subtype (α1, α2, β). Although α1-ARs were implicated in increased NA excitability, central adrenergic regions elevate heart rate (HR) and α1-AR agonism in CVNs increased inhibitory GABA receptor (GABAR) activity. Both these results are consistent with decreased (not increased) excitability. To clarify the role of α1-ARs in CVN-mediated cardiac regulation, we hypothesized α1A-ARs suppress CVN activity. Molecular biology confirmed expression of all α1-AR isoforms (A, B, D) in NA, including CVNs specifically. In urethane-anesthetized wild-type

(WT) mice, bilateral NA nanoinjection of the α1-AR agonist, phenylephrine (PE; 60 nL; 10 nL/s), produced a significant tachycardia (564.2±32.1 vs 625.7±20.9 BPM; n = 5; p = 0.03). In α1A-ARNull mice, PE elicited bradycardia (593.1±7.1 vs 555.5±8.7 BPM; n = 4; p = 0.01), and in mice lacking GABAR activity in NA (ChAT-δNull), PE induced a smaller, non-significant bradycardia (604.2±31.1 vs 591.4±39.7 BPM; n = 4). HR changes across genotypes differed significantly (WT: +61.3±39.3; α1A-ARNull: −37.6±13.0; ChAT-δNull: −12.2±26.1; one-way ANOVA, p = 0.001), with post hoc differences between WT and α1A-ARNull (p = 0.001) and WT and ChAT-δNull (p = 0.01). These findings indicate that activation specifically of α1A-ARs in NA increases HR (by decreased excitability) via increased inhibitory GABAR signaling. Future studies will use optogenetic stimulation of adrenergic terminals in NA and electrophysiological recordings to determine whether α1A-AR effects are direct or indirect. These studies provide new insight into and potential therapeutics to restore central circuit regulation of HR in CVD.

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Non-neuronal Tph1 ablation does not alter basal kidney function but mitigates ischemic kidney insufficiency in adult rats

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Abstract

The tryptophan hydroxylase (Tph) gene family encodes the rate-limiting enzymes for serotonin biosynthesis, a signaling molecule with diverse physiological and pathophysiological actions. Mammals express two isoforms: Tph1, producing peripheral non-neuronal serotonin, and Tph2, which drives neuronal serotonin synthesis. Both isoforms hydroxylate L-tryptophan to 5-hydroxytryptophan but have distinct biological roles. Tph1 is enriched in enterochromaffin cells and kidney proximal tubules, where serotonin modulates gut motility, secretion, vascular tone, and epithelial ion transport. Tph2 sustains serotonergic neurotransmission in the neurons. We investigated the contribution of peripheral serotonin to kidney function at baseline and during ischemia–reperfusion injury (IRI) using adult transgenic rats harboring a CRISPR-engineered 1-bp deletion in exon 4 of the Tph1 gene. Basal plasma and urinary L-tryptophan concentrations were unchanged in Tph1 knockout (KO) versus wild-type (WT) rats, whereas plasma serotonin and urinary 5-hydroxyindoleacetic acid were markedly reduced, confirming substantial loss of peripheral serotonin. Despite this deficit, mean arterial pressure and heart rate (telemetry), transdermal glomerular filtration rate, renal blood flow, renal vascular resistance, renal function indices, and kidney morphology were unchanged, indicating that non-neuronal serotonin is dispensable for maintaining resting renal hemodynamics and filtration in adult rats. Conversely, Tph1 ablation conferred significant protection during renal IRI. 35 minutes of renal ischemia and 24 hours of reperfusion increased renal Tph1 expression and serotonin production in WT kidneys, without altering intestinal Tph1 expression or serotonin levels, indicating a kidney-specific upregulation of serotonin in response to renal ischemic stress. KO rats exhibited improved renal perfusion, attenuated systemic hemodynamic instability, reduced tubular dilatation, cast formation, and necrosis compared to WT. These results identify Tph1-derived serotonin as a previously unrecognized mediator of renal ischemic insufficiency and implicate serotonin signaling in microvascular dysfunction and tubular injury that exacerbate ischemic acute kidney injury. Therefore, targeting peripheral serotonin may be a promising strategy for treating renal IRI.

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Interruption of LARP6-collagen mRNA association reduces pressure overload-induced cardiac dysfunction independent of fibrosis in female, but not male, mice

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Abstract

LARP (La ribonucleoprotein 6, Translational regulator) is a multifunctional RNA binding protein implicated in tissue fibrosis and dysfunction. Prior work in the liver has established that binding of LARP6 to a conserved 5' stem loop (5'SL) in collagen Iα chain mRNA increases collagen I translation and deposition. Despite being expressed in the heart, its role in cardiac remodeling and function are unclear. Therefore, we investigated the hypothesis that interruption of LARP6-collagen mRNA association prevents pressure overload (PO)-induced cardiac dysfunction and fibrosis. Both male and female mice with a knock-in mutation in the 5'SL region that prevents LARP6 association and wild-type (WT) littermate controls underwent PO by transverse aortic constriction, with sham-operated serving as controls. Over 4 weeks post-surgery, male, but not female, 5'SL mice had higher mortality than WT controls. Echocardiography revealed similar reduction in ejection fraction in male 5'SL mice compared to WT mice after PO while female 5'SL mice exhibited blunted reduction in ejection fraction compared to WT. Despite these functional changes, male and female 5'SL mice had similar cardiac hypertrophy and pulmonary edema in response to PO compared to WT mice. Furthermore, the cardiac fibrotic response to PO was not changed by sex or genotype. Bulk cardiac RNA sequencing of female hearts revealed dramatic transcriptomic changes associated with PO that were sensitive to 5'SL mutation. Specifically, the top gene networks in female 5'SL hearts after PO, versus WT mice, were enriched for genes associated with reduced cardiomyopathy and mitochondrial disorders as well as modulation of apoptosis and vasculogenesis. Together, these data suggest that interruption of the LARP6-collagen mRNA interaction lessens cardiac dysfunction induced by PO independent of cardiac fibrosis and in a sex-specific manner. This work provides rationale for interrogation of LARP6 binding partners and actions in the heart beyond its well-described fibrotic action in other tissues.

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A critical role of UCH-L1 in alleviating acute cardiorenal syndrome type-5 in sepsis

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Abstract

Sepsis is a systemic severe inflammatory condition that causes multi-organ dysfunction and has a mortality rate exceeding 25%. Cardiorenal syndrome type 5 (CRS-5) involves concurrent cardiac and renal dysfunction secondary to systemic insults such as sepsis. However, the molecular mechanisms underlying CRS-5 in sepsis and the contribution of CRS-5 to septic death remain poorly understood. Herein, we report a critical role of a deubiquitinating enzyme ubiquitin carboxyl-

terminal hydrolase L1 (UCH-L1) in the control of sepsis-induced CRS-5 and related death.

Using a lipopolysaccharide (LPS)-induced sepsis mouse model, cardiorenal dysfunction was confirmed by elevated circulating levels of troponin I, blood urea nitrogen (BUN), neutrophil gelatinase-associated lipocalin (NGAL) and a significant reduction in fractional shortening (FS) and stroke volume (SV) in wild type (WT) mice. Moreover, we found that UCH-L1 protein expression in the heart and kidney were correlated with cardiac and renal dysfunction. Interestingly, cardiac-restricted UCH-L1 (CR-UCH-L1) overexpression significantly reduced mortality, improved cardiac function as indicated by increased SV and FS, and alleviated renal injury as reflected by decreased BUN levels in the LPS-induced sepsis model. Notably, vascular cell adhesion molecule-1 (VCAM-1) expression was selectively reduced in both cardiac and renal tissues of CR-UCH-L1 overexpression mice compared with WT mice following LPS challenge, suggesting that cardiac UCH-L1 modulates cardiorenal inflammatory signaling pathways during sepsis. Together, these findings demonstrate that cardiac UCH-L1 plays a critical role in protecting against acute CRS-5 contributing to death in sepsis via a unique heart-kidney communication.

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Tissue-specific reduction of IL-10 and elevated inflammation in chronic intermittent hypoxia: Potential implications for obstructive sleep apnea

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Background

Obstructive sleep apnea (OSA) a chronic condition and is characterized by intermittent hypoxia (IH), which induces low-grade systemic inflammation that contributes to OSA-related morbidities. Interleukin-10 (IL-10), a critical anti-inflammatory cytokine, is reportedly unaffected by OSA when measured in plasma or serum. However, no studies have comprehensively assessed IL-10 levels across specific tissues in OSA. We hypothesized that IL-10 might be affected by IH in a tissue-specific manner.

Methods

Male C57Bl/6J mice were exposed to IH (FiO_2 cycling from 21% for 90s to 6% for 90s) or room air (RA, 21%) for 12 hours/day over six weeks. After euthanasia, levels of IL-10, interleukin-1 beta (IL-1β), and tumor necrosis factor-alpha (TNF-α) were measured in plasma, colon, visceral white adipose tissue (vWAT), and aortic tissue using ELISA.

Results

Plasma IL-10 levels did not differ between RA and IH groups (4.4 ± 2.4 pg/ml vs. 3.7 ± 2.3 pg/ml. p = 0.6). However, IH significantly reduced IL-10 levels in the colon (24.9 ± 1.8 pg/ml vs. RA: 14.4 ± 4.6 pg/ml, $p < 0.05$), vWAT (10.1 ± 0.9 pg/ml vs. RA: 4.4 ± 0.8 pg/ml, $p < 0.05$), and aorta (15.3 ± 1.3 pg/ml vs. 7.1 ± 2.4 pg/ml, $p < 0.05$). In contrast, plasma levels of IL-1β in mice exposed to IH were significantly higher when compared to control (3.9 ± 1.1 pg/ml vs. RA: 1.8 ± 0.5 pg/ml, $p < 0.05$) as well as colon (14.1 ± 1.7 pg/ml vs. RA: 5.1± 1.1 pg/ml, $p < 0.05$), vWAT (23.1 ± 4.1 pg/ml vs. RA: 2.6 ± 0.5 pg/ml, $p < 0.05$), and aorta (19.2 ± 4.5 pg/ml vs. RA: 4.2 ± 0.8 pg/ml, $p < 0.05$). Similarly, TNF-α levels were significantly elevated in IH-exposed mice plasma (7.1 ± 2.1 pg/ml vs. 2.3 ± 0.6 pg/ml, $p < 0.05$), colon (44.7 ± 3.7 pg/ml vs 21.4 ± 2.1, $p < 0.05$), vWAT (34.4 ± 5.7 pg/ml vs. 11.4 ± 1.9 pg/ml, $p < 0.05$), and aorta (31.3 ± 4.1 pg/ml vs. 14.3 ± 2.9 pg/ml, $p < 0.05$).

Conclusion

IH exposures mimicking OSA reduce IL-10 levels in tissues but not in plasma, while concurrently increasing pro-inflammatory cytokines IL-1β and TNF-α in both plasma and tissues. These results suggest that IH induces a tissue-specific inflammatory imbalance, characterized by reduced anti-inflammatory responses and heightened pro-inflammatory signaling.

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α1A-Adrenergic Receptor Expression on Macrophages Elicits a Proinflammatory Response During Heart Failure Progression

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Abstract

Heart failure (HF) is the leading cause of death worldwide and inflammation is a key regulator of HF progression. A major regulator of immune response are adrenergic receptors (AR), which mediate the cellular effects of the sympathetic nervous system (SNS). The β2AR subtype is widely expressed on immune cells where it has been shown to modulate a variety of responses while other subtype expression is restricted, and their role is poorly defined.

Macrophages are vital immunoregulatory cells that influence inflammation based on phenotype. Preliminary findings from our laboratory demonstrates that macrophages upregulate α1A-AR expression under proinflammatory conditions. We hypothesized α1A-AR elicits proinflammatory responses and immune specific deletion of α1A-AR would reduce proinflammatory responses in HF. To examine the effect of α1A-AR expression on macrophages, lipopolysaccride was applied to WT and α1A-AR knockout (KO) bone marrow-derived macrophages (BMDM). α1A-AR expression amplified pro-inflammatory cytokine production, while KO reduced responses. To understand whether the proinflammatory phenotype of α1A-AR in macrophages exists in vivo, a chronic isoproterenol (ISO) model of HF was used to mimic the prolonged SNS overactivation that occurs in HF. Mice were irradiated to deplete endogenous bone marrow (BM) and received WT or α1A-ARKO BM transplant (BMT). WT BMT control mice receiving ISO exhibited hallmarks of HF, including increased hypertrophy and fibrosis, which were blunted in α1A-ARKO BMT animals. This corresponded with worsened functional parameters in WT BMT mice, which were improved with α1A-ARKO BMT animals. These findings demonstrate that mice that immune cell α1A-ARKO improves outcomes in a chronic βAR activation model of HF. These findings are significant in that they are the first to show an upregulation of α1A-AR during proinflammatory conditions, which impacts HF development and progression and represent a inflammatory pathway which may be a novel therapeutic target for the treatment of HF.

Therapy development for X-linked deletion and expansion diseases in females

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Abstract

Dosage imbalance of X-linked genes is a central driver in multiple human conditions, contributing to both neurodevelopmental and cardiovascular symptoms. Neurologically, loss of FMR1 gene expression underlies Fragile X Syndrome, the most common inherited cause of intellectual disability and autism. Cardiovascular manifestations are also prominent in X-linked disorders, particularly in Turner syndrome, where haploinsufficiency of X-linked genes contributes to congenital heart disease, vascular dysfunction and aortic abnormalities. Despite these clinical links, strategies to restore dosage of X-linked genes are not studied and no approved drugs are available.

X-chromosome inactivation (XCI) further complicates disease severity in females, as cellular mosaicism and skewed XCI patterns can lead to

variable expression of disease-causing alleles. When the active X carries a deletion or silenced gene, the inability to access the intact allele on the inactive X exacerbates functional gene loss. This creates an opportunity to therapeutically target the inactive X (Xi) to restore gene dosage without introducing new genetic alterations.

This project explores targeted reactivation of genes on the inactive X chromosome (Xi) as a therapeutic strategy to compensate for X-linked gene loss. We focus on two complementary molecular approaches: (i) antisense oligonucleotide (ASO) mediated knockdown of XIST, a key regulator of Xi, and (ii) transcriptional activation using dCas9-VPR. Together, these strategies aim to weaken global Xi and directly enhance transcription of X-linked genes such as FMR1.

Initial studies established that X-linked genes show measurable transcriptional responsiveness to XIST modulation, supporting feasibility of this approach. Further studies test female patient-derived iPSCs as relevant models of gene-dosage regulation. Using multiple patients and isogenic iPSC clones only differing in Xi, this platform enables systematic evaluation of X-linked gene reactivation strategies.

Ultimately, this will test whether X-linked gene dosage modulation can stabilize or restore neural and cardiovascular functions, advancing therapies for X-linked disorders.

Chemotherapy versus radiation for cardiac tumors

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Background

Malignant cardiac tumors (MCT) pose substantial treatment challenges due to their close proximity to critical cardiac structures and the common occurrence of delayed diagnosis. Standard treatment approaches often involve chemotherapy and radiotherapy. This study aims to compare the treatment outcomes of these two modalities in patients diagnosed with MCT.

Methods

We reviewed data on 783 patients diagnosed with MCTs from the Surveillance, Epidemiology, and End Results (SEER) program from 2000 to 2021. Survival distribution trends were calculated for variables such as radiation type and chemotherapy type using Kaplan-Meier analysis and Cox Proportional Hazards Regression.

Results

We found a significant difference in survival between the chemotherapy and radiotherapy groups, with chemotherapy showing better outcomes ($p = 0.006$). Most common tumor types were hemangiosarcoma, diffuse large B cell lymphoma and giant cell sarcoma. Chemotherapy had a higher survival rate (16%) than beam (11%) and general radiation (13%). Over 5 years, chemotherapy had the highest survival rates. Notably, no patients received combined treatment.

Conclusion

This study highlights the higher survival rates in MCT patients treated with chemotherapy. These findings underscore the importance of further research to optimize chemotherapeutic treatment strategies for MCT patients. It also opens the question on utility of combined chemo and radiotherapy in highly malignant tumors.

Effect of High Protein Intake on Endothelial Function

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Abstract

Endothelial dysfunction is an early marker of cardiovascular disease and is strongly influenced by dietary factors. Although high protein intake (HPI)

is commonly recommended for weight management and metabolic health, its effects on vascular function and underlying mechanisms remain incompletely understood. We hypothesized that HPI impairs endothelial function through postprandial elevations in circulating amino acids and suppression of fibroblast growth factor-21 (FGF-21) signaling.

In a randomized crossover study, participants consumed isocaloric standard-protein or high-protein mixed meals. Postprandial endothelial function was assessed using the reactive hyperemia index (RHI), and plasma amino acid and FGF-21 concentrations were measured before and for 150 minutes after starting the meal. To evaluate the effects of increasing amounts of amino acids and FGF-21 on endothelial cells, we conducted in vitro experiments with human umbilical vein endothelial cells (HUVECs) and mouse vascular tissue.

High protein intake increased postprandial plasma amino acid concentrations and reduced circulating FGF-21, accompanied by a decrease in RHI. In endothelial cells, amino acid exposure increased reactive oxygen species production, reduced stimulatory eNOS phosphorylation (Ser1177), increased inhibitory eNOS phosphorylation (Thr495), and decreased nitric oxide production. Amino acid treatment also increased inflammatory gene expression and vascular inflammasome activation in mouse arteries. In contrast, FGF-21 treatment enhanced eNOS activation and nitric oxide production in a dose-dependent manner.

These findings suggest that high protein intake impairs endothelial function, likely mediated by amino acid–induced oxidative and inflammatory stress, as well as the suppression of FGF-21 signaling.

Sex-specific myocardial dysfunction in cardiometabolic HFpEF

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Introduction

HFpEF (Heart Failure with preserved Ejection Fraction) is dominated by the impaired diastolic function, and accounts for ~50% of all heart failure cases. HFpEF disproportionately affects women, with prevalence increasing sharply after menopause. Clinical studies suggest that HFpEF in men and women are distinct, understanding sex-specific pathophysiology in HFpEF is essential for achieving an in-depth understanding of HFpEF and developing personalized therapeutic strategies. Hypothesis: Sex and hormone status differentially affect myofilament mechanical properties, supporting the need for sex-specific HFpEF therapeutic interventions.

Methods

Menopause was induced in female mice using 4-vinylcyclohexene dieopxide (VCD). Then HFpEF conditions were induced in both male and postmenopausal female mice using a 2-hit regimen (high-fat diet plus LNAME-supplement) for 4 months. We used a myocardial slice mechanical technique to assess the cardiac function at the intact myocardial levels. Left Ventricular (LV) slices (200 μm thickness) were hooked between a force transducer and the piezo length controller. The piezo movement was controlled to generate force-length loops. Key loop parameters were assessed, including end-diastolic stress-length relationship (EDSLR) as a measure of passive myocardial stiffness, isometric relaxation time (IMRT) as an index of relaxation kinetics, stroke length (equivalent to stroke volume), and stroke work (mechanical output). The relative contributions of microtubules, diastolic crossbridge, and titin were assessed using colchicine and 2,3-butanedione monoxime (BDM).

Results

Preliminary findings indicate that the female-HFpEF-like (VCD-2hit) group showed a trend toward increased IMRT and EDSLR, suggesting delayed relaxation increased LV diastolic stiffness. Stroke work and length decrease, suggesting reduced cardiac output. Additional male cohort is currently being included.

Conclusion

Cardiac slice mechanical technique is a useful tool for studying myocardial mechanics in intact cardiac tissue. Preliminary result suggests that the female-2hit-VCD mice develop HFpEF-like condition under metabolic stress, observed as increased myocardial stiffness and delayed relaxation.

Role of CD4+ β2-Adrenergic Receptor Signaling in Cardiac Remodeling in a Pressure-Overload Model of Heart Failure

Background

Heart failure often leads to pathological cardiac remodeling, significantly influenced by immune mechanisms. CD4+ T cells, through β2 adrenergic receptor (β2AR) signaling, are hypothesized to play a crucial role in this process. The impact of β2AR signaling in CD4+ T cells on cardiac remodeling and fibrosis in heart failure conditions remains to be fully elucidated. Objective: This study investigates the role of CD4+ β2AR signaling in cardiac remodeling under pressure-overload conditions, hypothesizing that deletion of β2AR in CD4+ T cells would impair the remodeling process.

Methods

We utilized a murine model of heart failure induced by transverse aortic constriction (TAC) to study the effects of β2AR signaling in CD4+ T cells. Wild-type mice were compared to CD4+ T cell-specific β2AR knockout mice(tKO). Cardiac function was assessed via echocardiography, fibrosis was evaluated through histological analysis, and inflammatory responses were measured using molecular assays.

Results

The deletion of β2AR in CD4+ T cells led to a marked reduction in cardiac remodeling and fibrosis, indicating that β2AR signaling in these cells is critical in driving the pathological changes observed in pressure-overload heart failure. The results showed that tKO mice exhibited less fibrosis and a reduced inflammatory response, including lower ICAM-1 gene expression, highlighting the significant impact of β2AR signaling on these processes

Conclusion

Our findings demonstrate that β2AR signaling in CD4+ T cells plays a significant role in cardiac remodeling and fibrosis during heart failure. The absence of this signaling dampens these processes, suggesting that modulating β2AR activity in CD4+ T cells could be a potential therapeutic strategy to mitigate adverse cardiac remodeling.

Divergent Synaptic Adaptations in GABAergic vs. Glutamatergic nTS Pathways May Underlie Respiratory Responses to Sustained Hypoxia

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Abstract

Exposure to hypoxia occurs during physiological (e.g., high altitudes) and pathophysiological (e.g., obstructive pulmonary disease) conditions. The nucleus tractus solitarii (nTS) integrates sensory input from hypoxia-sensitive peripheral chemoreceptors, which release glutamate to initiate reflex responses. Yet, the magnitude of these responses and overall nTS activity is determined through a balance of glutamate (Glu) and gamma-aminobutyric acid (GABA) signaling. This study investigated how sustained hypoxia (SH) affects GABA neurons in the nTS and cardiorespiratory function.

Male and female GAD1-EGFP mice (4-5 weeks) were used to differentiate GABA (GAD+) and presumptive Glu (GAD-) neurons in the nTS. Mice were exposed to SH (10% O2, 24 hours) or normoxia (21% O2, 24 hours) before experimentation.

Plethysmography revealed increased tidal volume and minute ventilation following SH. Immunohistochemistry showed increased c-Fos in the nTS after SH, primarily in Glu neurons, suggesting increased nTS Glu activity. Whole-cell patch-clamp recordings in nTS slices examined synaptic neurotransmission and electrophysiological properties of GAD+ and GAD- neurons. Neurons were characterized by mono- or polysynaptic connection with the tractus solitarii (TS) via TS stimulation. Under normoxic conditions, GAD+ and GAD- neurons had similar properties with notable exceptions: monosynaptic GAD+ neurons had greater spontaneous excitatory postsynaptic current (sEPSC) frequency than GAD- neurons, while polysynaptic GAD+ neurons showed decreased synaptic throughput during repetitive (20 Hz) TS stimulation. Following SH, monosynaptic GAD- and GAD+ neurons became more depolarized without altered synaptic properties. Conversely, SH differentially affected polysynaptic neurons, altering presynaptic release properties and enhancing GAD+ action potential discharge. SH increased synaptic throughput in both polysynaptic phenotypes at 20 Hz TS stimulation.

These data demonstrate that SH drives differential synaptic remodeling of nTS circuitry by altering transmission and excitability of GABAergic and glutamatergic neurons, possibly contributing to observed respiratory changes.

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CYLD Suppresses VSMC Phenotypic Switching into Synthetic VSMCs for Neointimal Formation by Restricting HIF-1α–Dependent Metabolic Reprogramming

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Abstract

Vascular smooth muscle cell (VSMC) phenotypic switching is central to the development and progression of vascular diseases, such as restenosis and atherosclerosis. However, the molecular mechanisms driving this process remain incompletely understood. Here, we sought to determine the role of a deubiquitinating enzyme cylindromatosis (CYLD) in VSMC phenotypic switching into synthetic VSMCs for occlusive lesion formation in the artery. Under physiological conditions, we found a low level of CYLD expression in the carotid artery primarily localized to VSMCs in the tunica media. After injury, CYLD expression was markedly increased, concomitant with VSMC phenotypic switching into synthetic VSMCs characterized by a reduction in the expression of contractile VSMC markers and an increase in the expression of secretory proteins as well as neointima (NI) formation. Notably, NI formation was significantly enhanced in CYLD knockout (KO) mice, accompanied by increased VSMC proliferation in the injured arteries. Lineage tracing analyses further revealed that CYLD KO promoted phenotypic switching of pre-existing mature VSMCs into synthetic VSMCs and their expansion, leading to increased NI formation in injured arteries. Consistent with these in vivo findings, in vitro studies showed that CYLD deficiency promoted VSMC proliferation and migration, along with a transition from the contractile to the synthetic phenotype. RNA sequencing analysis revealed that CYLD deficiency enhanced HIF-1α–driven transcription of glycolytic genes and a metabolic shift from mitochondrial oxidation toward glycolysis, thereby facilitating VSMC growth and phenotypic switching from the contractile to the synthetic phenotype. Collectively, these findings identify CYLD as a critical suppressor of VSMC phenotypic switching into synthetic VSMCs for neointimal formation in injured arteries, through inhibition of HIF-1α–dependent metabolic reprogramming.

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