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学位論文題目	Nicotinamide Phosphoribosyltransferase (Nampt) /Nicotinamide Adenine Dinucleotide (NAD) Axis Suppresses Atrial Fibrillation by Modulating the Calcium Handling Pathway(ニコチンアミドホスホリボシルトランスフェラーゼ(Nampt)/ニコチンアミドアデニンジヌクレオチド(NAD)軸はカルシウムハンドリングを調節することによって心房細動を抑制する)
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Abstract of thesis

In this doctoral dissertation, Feng Duo describes the pathophysiology of Atrial Fibrillations (AF). The content is summarized as follows.

Purpose

AF is the most common chronic cardiac disease. Also, AF is a quivering or arrhythmia that can lead to blood clots, stroke, heart failure and other heart-related complications. Nicotinamide phosphoribosyltransferase (Nampt) is the rate-limiting enzyme that catalyzes nicotinamide adenine dinucleotide (NAD) activity. Nampt and NAD are essential for maintenance of cellular redox homeostasis and modulation of cellular metabolism, and their expression levels decrease with aging and obesity. However, a role for Nampt in AF is unknown. The author aimed to test whether there is a role of Nampt/NAD axis in the pathogenesis of obesity-induced AF.

The author also aimed to explore the effects and mechanisms of how Nampt affects AF occurrences by a mouse AF model using both wild-type (WT) mice and heterozygous Nampt knockout (NKO) mice.

Material and method

Male C57BL/6J (WT) mice and heterozygous NKO mice were fed with a normal chow diet (ND) or a high-fat diet (HFD). At 5 and 13 weeks of age, parasternal long-axis and short-axis views were obtained at the papillary muscle level under anesthesia with isoflurane using an echocardiographic system. The heart rate (HR), left ventricular end-diastolic diameter (LVDd), left ventricular end-systolic diameter (LVD's), fractional shortening (FS), left ventricular ejection fraction (LVEF), left atrial dimension (LAD), and left ventricular systolic volume (LVV) were determined. LVEF was calculated by the Teichholz method. The author isolated adult atrial cardiomyocytes from WT and NKO mice atriums using a Langendorff-free procedure. Live cell imaging was performed with a 40x lens on a TCS SP5 Confocal Microscope System.Calcium sparks were detected and characterized following established criteria. To evaluate calcium sparks, cytoplasmic fluorescence signals were obtained using LAS-X software. To evaluate Nampt expression, we performed immunohistochemistry. After deparaffinization and antigen activation, the sections were incubated with a rabbit anti-Nampt monoclonal antibody. All data are expressed as mean ± standard error of the mean. For comparisons between the groups, continuous values were analyzed by one-way ANOVA followed by a post-hoc Bonferroni test.

Results

There were no significant differences in echocardiographic parameters and atrial fibrosis between WT and NKO mice. HFD and NKO significantly decreased the Nampt expression and NAD levels. AF inducibility was significantly increased in WT+HFD, NKO+ND, and NKO+HFD mice compared with WT+ND mice. AF duration was significantly longer in WT+HFD and NKO+ND mice and further prolonged in NKO+HFD mice compared with WT+ND mice. Expression levels of oxidized CaMKII and phosphorylated RyR2 were increased in NKO+HFD mice, and diastolic calcium leaks from the sarcoplasmic reticulum were facilitated in these mice. Treatment with nicotinamide riboside, a NAD precursor, partially restored the HFD-induced AF perpetuation.

Discussion

The author revealed HFD feeding and heterozygous NKO prolonged catheter-induced AF duration without significant structural abnormalities. HFD and NKO enhanced the expressions of oxidized CaMKII and phosphorylated RyR2 and induced frequent diastolic Ca2+ leaks in cardiomyocytes. Furthermore, NAD precursor NR partially rescued the arrhythmogenic phenotypes. It is suggested that the Nampt/NAD axis plays a protective role in AF pathogenesis by regulating the calcium-handling pathway.

Abstract of assessment result

(General Comments)

The author revealed that heterozygous NKO directly enhanced oxidized CaMKII-mediated phosphorylation of RyR2, and diastolic Ca2+ leaks from the SR in cardiomyocytes, leading to facilitation of AF in HFD-fed mice. Supplementation with NAD precursor NR partially rescued the high AF Vulnerability. Therefore, the Nampt/NAD axis may be a novel therapeutic Target for AF treatment. The final examination committee conducted a meeting as a final examination on Nov.17, 2020. The applicant provided an overview of dissertation, addressed questions and comments raised during Q&A session. All of the committee members reached a final decision that the applicant has passed the final examination. The final examination committee approved that the applicant is qualified to be awarded Doctor of Philosophy in Medical Sciences.