The mechanism of βAR stimulation-induced IL-6 production in cardiac fibroblast

Shota Tanaka, Kota Tonegawa, Shota Fuchigami, Atsuki Imaeda, Masanori Obana, Makiko Maeda, Yasushi Fujio, Hiroyuki Nakayama

Laboratory of Clinical Science and Biomedicine, Graduate School of Pharmacological Sciences, Osaka University, Japan

Background and Objective: Inflammation occurs in the damaged heart and β adrenergic receptor (βAR) signaling could play an important role in cardiac inflammation. Cardiac fibroblasts (CFs) are one of the cardiac composing cells, and various stimuli including βAR activation promote cytokine production in CFs. Although CFs are involved in the development of heart failure via cytokine production, its molecular mechanism remains unclear. Therefore, we investigated to determine how βAR stimulation induces proinflammatory cytokines in CFs and to identify the new therapeutic targets preventing cardiac inflammation.

Methods and Results: CFs were isolated from adult wild-type mice and the expression of proinflammatory cytokines, namely, IL-6, IL-1β and TNFα, was assessed by quantitative RT-PCR after βAR stimulation with isoproterenol (ISO, 1 or 10 μM). As a result, IL-6 mRNA was upregulated 13-fold compared to basal condition 1 hour after ISO treatment. However, mRNA expression of IL-1β and TNFα showed little change after ISO treatment. Forskolin, in response to βAR-independent adenylyl cyclase activator, the mRNA expression of IL-6, but not that of IL-1β or TNFα, was increased in a dose-dependent manner. Pretreatment with H-89, 5Z-7-Oxozeaenol or BAY11-7082 as PKA, TAK1 or NF-κB inhibitor, respectively, suppressed ISO-induced IL-6 upregulation. In contrast, N-acetylcysteine, an anti-oxidant, failed to reduce IL-6 mRNA increase. Furthermore, immunoblot analyses revealed that IL-1β-mediated induction of IL-6 was associated with activation of p-38 MAPK and JNK, while ISO did not activate these signaling pathway. Interestingly, the mRNA expression of Arid5a, an IL-6 mRNA stabilizer, showed 4-fold increase after 1 hour ISO treatment and suppressed its upregulation by the pretreatment with H-89, 5Z-7-Oxozeaenol or BAY11-7082. Finally, shRNA suppressing Arid5a expression decreased IL-6 mRNA induced by ISO treatment.

Conclusion: In response to βAR stimulation, CFs produced IL-6 through Arid5a mainly by activating adenylyl cyclase, PKA, TAK1 and NF-κB as a novel proinflammatory response in the heart.