24. Cardiomyocytes Produces Acetylcholine in Response to Muscarinic Receptor Stimulation: a Possible Mechanism for Amplification of Cardioprotective Effects of Cardiac Vagal Nerve

Kakinuma Y, [1]Akiyama T, Arikawa M, Handa T, Sato T

Dep. of Cardiovascular Control, Kochi Medical School, Nankoku, Japan.
[1]Dep. of Cardiac Physiology, National Cardiovascular Center Research Institute, Osaka, Japan

Background: Our previous studies have demonstrated that vagal stimulation (VS) increases the survival of rats with chronic heart failure, and acetylcholine (ACh) plays a protective role in cardiomyocytes against prolonged hypoxia through a non-hypoxic induction pathway of PI3K/Akt/HIF-1α, and finally an acetylcholinesterase inhibitor, donepezil, an anti-Alzheimer's disease drug, also prevents a progression of heart failure in rats with myocardial infarction. However, taken with evidence that vagal innervation is extremely poor in the left ventricle, there is a missing link between VS, ACh and donepezil, suggesting another source of ACh exists in the heart.
Hypothesis: We assessed the hypothesis that ACh is produced by cardiomyocytes and ACh promotes its synthesis via a positive feedback system, which donepezil could modulate.
Methods and Results: Rat primary cardiomyocytes expressed choline acetyltransferase (ChAT), a crucial enzyme for ACh synthesis and its signal was localized in cytoplasm. Vesicular ACh transporter (VAChT) was also detected in cytoplasm with the vesicular structure identified by immunogold electron microscopy, suggesting that cardiomyocytes possess crucial components for ACh synthesis. Intracellular ACh in rat cardiomyocytes was detected not only with physostigmine (5.2 ± 0.4 nM in cell lysates), but also with donepezil, both of which are acetylcholinesterase inhibitors. In response to exogenous ACh or pilocarpine, a muscarinic agonist, cardiomyocytes significantly increased the transcriptional activity of ChAT and ChAT protein expression, and consequently the intracellular ACh level was up-regulated to 255 ± 28 % ($p < 0.05$) by pilocarpine, suggesting an ACh-amplification mechanism. Surprisingly, donepezil elevated the ChAT prompter activity and ChAT protein expression, leading to an increase in the ACh level to 385 ± 103% ($p < 0.05$) in cardiomyocytes in vitro.
Conclusion: The present results suggest that cardiomyocytes have an ACh synthesis system that acts through a muscarinic receptor, which is positively modulated by cholinergic agents including donepezil. Such an ACh-amplification system in cardiomyocytes may be involved in the beneficial effects of VS on the cardiac ventricles.