Atrial fibrillation (AF) is associated with reduced L-type Ca\textsuperscript{2+} current (ICa,L) and altered other forms of heart failure (HF). Mechanisms accounting for these beneficial effects remain however only partially understood. In particular, due to the difference in receptor targeting and to the great variability in human HF-patients response, the specific mechanism of action of β-blockers is still under investigation. We have recently demonstrated, in an animal model of HF, that a reciprocal down-regulation occurs between βAR and the cardioprotective sphingosine-1-phosphate (S1P) receptor-1 (S1PR1). Purpose: Hence, we hypothesize that, in addition to salutary actions due to direct βAR blockade, agents such as metoprolol improve post-MI structural and functional outcomes via restored protective S1PR1 signal, and we sought to determine mechanisms accounting for this effect.

Methods and Results: In HEK293 cells and in vitro cardiomyocytes, metoprolol (Meto) prevented isoproterenol (βAR agonist)-dependent S1PR1 down-regulation. Treatment of infarcted mice with Meto or S1P (one week after MI for 3 weeks) markedly ameliorated cardiac function and prevented remodelling, while preserving cardiac plasma membrane S1PR1 whose levels were down-regulated in untreated MI mice. Next, we co-infused infarcted mice with S1P and Meto, and found no additional beneficial effects. Since previous evidence attests that Meto can increase cardiac βARs levels and activity, and this receptor in adipocytes is responsible for S1P secretion, we measured basal and Meto-stimulated cardiac Sphingosine kinase 1 (SphK1), the enzyme responsible for S1P secretion, and circulating S1P levels in MI, cAF and dβAR KO mice. These animals displayed markedly reduced levels of both, not rescued by Meto. Importantly, the βAR blocker did not ameliorate post-MI dysfuntion in βAR KO mice.

Conclusions: βAR-blockers enhance βAR-signaling, promoting the secretion of S1P that, in turn, activates the S1PR1 signaling. These signaling interactions represent a previously unrecognized mechanism whereby βAR blockers prevent post-MI decompensation and adverse remodelling.

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B-blocker therapy in heart failure reduces migratory and proliferative properties of primarily cultured failing cardiac fibroblasts via reduction of g protein-coupled receptor kinase-2 expression

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Introduction: Cardiac remodeling is a cardinal process mediating the progression of heart failure (HF). Cardiac fibroblasts (CFs) play a critical role in the regulation of left ventricular remodeling following myocardial infarction (MI), mainly through differentiation to a myofibroblast phenotype that expresses increased migratory and proliferative properties. Among several therapeutic effects of β-blocker on cardiac function, this class of drugs is known to slow the process of cardiac remodeling and to reduce cardiac fibrosis. G protein-coupled receptor kinase-2 (GRK2) is involved in the mechanism of maladaptive ventricular remodeling and regulates CF function. Indeed, this kinase has been shown to be upregulated and to induce βAR uncoupling in CFs extracted from human failing hearts.

Purpose: The aim of the present study was to evaluate the effects of β-blocker therapy on CF activation and on GRK2 levels in HF.

Methods and Results: MI was surgically induced in mice and, 4 weeks later, mice were randomized to receive either Metoprolol (Meto) or vehicle for 4 additional weeks. Sham-operated mice were also included. Echocardiography, performed after 4 weeks of treatment, revealed that Meto was able to prevent cardiac functional deterioration and remodeling observed in HF control mice. Moreover, β-blocker therapy resulted in reduced cardiac apoptosis and fibrosis. At the end of the study period, GRK2 was markedly increased in CFs extracted from HF mice compared to those extracted from sham. Importantly, Mет treatment significantly limited HF-related GRK2 upregulation in CFs. Next, we assessed proliferation and migration in primarily cultured CFs extracted from the 3 study groups. Proliferation and migration were robustly increased in CFs extracted from HF controls compared to sham. Mет treatment resulted in a normalization of both proliferation and migration of failing CFs. Furthermore, we extracted CF plasma membranes obtained from untreated failing hearts and we observed a robust βAR down-regulation compared to CFs extracted from sham. Importantly, Mет treatment resulted in a complete restoration of HF-related βAR dysfunction observed in failing CFs. Finally, in vitro overexpression of GRK2 in Mет-treated failing CFs completely abolished the beneficial effects of β-blocker therapy on CF, recapitulating a HF phenotype with increased proliferaton and migration.

Conclusions: GRK2 appears to have a relevant role in regulating CF proliferation and migration and reduced expression of this kinase may be an important mechanism of the positive effects of β-blocker therapy on HF-related maladaptive remodeling mediated by CFs.