Tenascin-C deficiency improves cardiac and vascular function in diabetic mice

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Background and aims: Diabetic cardiomyopathy (CMP) is known for cardiac and vascular dysfunction in lack of structural heart disease. Recently, Tenascin-C (TNC) upregulation in the myocardium and serum has been linked to worse outcome in diabetes and heart failure. However, the role of TNC in the development of diabetic CMP is not known. We aimed to study the function of TNC in the progression of cardiovascular dysfunction in diabetes. **Materials and methods:** AJ and TNC-KO adult male mice were repeatedly injected with streptozotocin (50mg/kg) to induce diabetes. Cardiac function was measured by echocardiography at baseline and at 18-20 weeks. Vascular endothelial function was assessed by wire myography in isolated thoracic aorta segments. Cardiac fibrosis and coronary network geometry were assessed. In addition, the effects of purified human TNC (phTNC) on isolated working rat hearts were evaluated. To clarify the source of TNC, a cellular model of diabetic CMP using human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) was established. Moreover, human ventricular cardiac fibroblasts (HCF) were cultured, then starvated and treated with TGF-β; phTNC (10µg/ml) and TLR4 inhibitor in combination with TNC and subsequently mRNA expression of α-SMA, TNC, Col-1, Col-3 and ACE1 were assessed by RT-qPCR. Finally, human umbilical vein endothelial cells (HUVEC) were treated either with phTNC (10µg/ml) or combination with TLR-4 inhibitor (TAK-242, 50nM) and analysed the expression of NADPH oxidase 1 and 4 (NOX1, NOX4), and interleukin-6 (IL-6). **Results:** TNC deficiency was accompanied by preserved ejection fraction and endothelium-dependent relaxation ($p<0.05$ and $p<0.001$, respectively). Histology revealed less cardiac fibrosis in TNC-KO diabetic animals than in the AJ diabetic group ($p<0.01$). Larger coronaries showed multiple branching distally and thicker walls in diabetic animals, while TNC-KO diabetic mice had richer branching systems, suggesting better left ventricular (LV) perfusion. In addition, cumulative dosage of phTN-C (80 ng/ml) resulted in a significant reduction in cardiac output ($p<0.01$) and LV systolic pressure ($p<0.05$) in isolated rat hearts. HiPSC-CMs under diabetic conditions did not upregulate TNC. In contrast, TGF-β treatment upregulated TNC expression in HCF ($p<0.01$). Notably, HCF exposed to phTNC promoted both α-SMA and ACE1 mRNA expression ($p<0.05$). In addition, HUVEC incubated with phTNC showed increased expression of IL-6 and oxidative stress-related markers (NOX4) while TLR-4 inhibitor pre-treatment markedly reversed these changes. **Conclusion:** TNC creates a fibrosis and oxidative stress facilitating environment, which leads to cardiomyocyte and endothelial cell dysfunction. Thus, TNC may be a critical mediator as well as a potential therapeutical target in the progression of cardiovascular dysfunction in diabetes.

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