Effects of Diabetic Conditions on Cardiac Fibroblasts

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March 31st, 2022, at 3:30 PM EST in Rhodes Annex 111

Introduction: Diabetic cardiomyopathy is a distinct pathophysiological condition that occurs in patients with diabetes mellitus and manifests in the absence of other comorbidities, such as coronary artery disease, hypertension, and myocardial infarction. Congestive heart failure is a condition characterized by the heart's loss of ability to adequately pump blood to the body. Over six million Americans have heart failure. Congestive heart failure can occur with a chronic condition or can have an acute onset. A particularly common chronic etiology of congestive heart failure is diabetes mellitus. 14.5% of type one diabetes mellitus (T1DM) patients and 35% of type two diabetes mellitus (T2DM) patients have some cardiac dysfunction.[3]

Cardiac fibroblasts (CFs) play a critical role in cardiac function. CFs synthesize the extracellular matrix (ECM) in the heart and differentiate into myofibroblasts after a cardiac injury. In a non-diabetic, healthy patient, CFs primarily work to maintain homeostasis. CFs synthesize collagen, elastin, bioactive molecules (TNFα, AngII, VEGF, etc.), and remodeling enzymes, such as matrix metalloproteinases (MMPs). MMPs break down the collagen and ECM to help maintain homeostasis in normal patients. In diabetic patients, hyperglycemia produces advanced glycation end products (AGEs) in the ECM. In addition, the ECM is more crosslinked, and therefore stiffer in diabetic patients. As a result, MMPs cannot bind to and break down the collagen.[1] Cardiac metabolism is also affected by the hyperglycemia associated with diabetes mellitus. Under normal physiological conditions, cardiomyocytes favor free fatty acid metabolism over glucose metabolism, but can alternate based on need.[2] In diabetic patients, cardiomyocytes increase the intake of fatty acids and β-oxidation to maintain a proper level of ATP production. Eventually, the cells cannot metabolize all the free fatty acids and the overaccumulation causes lipotoxicity, resulting in hypertrophy and the production of reactive oxygen species and reactive nitrogen species.[3] As a result, cardiomyocyte death occurs, leading to a profibrotic response and the upregulation of fibrosis.[4] Fibrosis and ECM remodeling cause increased ventricular stiffness, thus reducing ejection fraction and leading to congestive heart failure.

Objective: The primary objective of this project is to develop an in vitro model to ascertain how diabetes mellitus affects cardiac fibroblasts. A 3D printed tissue scaffold will be seeded with cardiac fibroblasts and exposed to different concentrations of glucose to model diabetes mellitus. An ELISA assay will be run to quantify the upregulation and downregulation of certain proteins of interest.

Aim 1: Manufacture a 3D printable hydrogel to create 3D cultures of cardiac fibroblasts.
Based on previous research, a protocol has been previously developed to create a hydrogel from porcine ECM. We hypothesize that this hydrogel could be crosslinked to make it compatible with Cellink's bioprinter. 3D printed tissue scaffolds will be printed on to Flex cell plates to mechanically condition them and will be seeded with cardiac fibroblasts.

Aim 2: Determine the effects of diabetes mellitus on cardiac fibroblasts.
We hypothesize that there will be a difference in levels of expression of α2β1, CDH2, CDH12, Angiotensin II, αSMA, and 6TLR in cardiac fibroblasts exposed to hyperglycemic conditions.

References:

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