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Platelet-Rich Plasma Enriched Cardiac Extracellular Matrix Hydrogel Increases Metabolic Activity and Induces Differentiation of H9c2 Cardiomyoblast

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Extended Abstract

A significant amount of research is being directed towards developing alternative therapies to repair the damaged heart muscle [1]. Cardiac extracellular matrix (cECM) hydrogel is a prospective tool as an injectable therapy to be explored as a minimally invasive procedure for cardiac tissue repair. The use of hydrogels becomes more advantageous as it allows the incorporation of molecules of interest. Our group has combined biomaterials with platelet-rich plasma (PRP), due to its easily endogenous accessible source of growth factors. In skeletal muscle repair, PRP therapies impact regeneration by directly influencing myoblast cells, which activities are controlled by growth factors [2]. Once cardiomyocytes do not undergo mitoses, the stimulation of cardiac myoblasts could assist cardiac tissue repair. Considering that, we aimed to develop a PRP-cECM hydrogel, to produce an enriched hydrogel that can deliver bioactive molecules and facilitate the regeneration of cardiac tissue. For this, myocardium from porcine ventricles was minced and decellularized in a 1% wt/vol sodium dodecyl sulfate (SDS) in 1× phosphate buffered saline (PBS) for 5 days, extensively washed with distilled water, frozen and freeze dried. Following, the scaffold was enzymatically digested with pepsin and its pH adjusted to 7.4. Then, activated PRP was incorporated into the gel in two concentrations: 10% and 20% (v/v). Both materials (decellularized scaffold and hydrogels) were analysed to assess residual DNA, histological and histochemical analysis, SEM, FTIR, collagen content and cytotoxicity and metabolic activity assays in H9c2 cardiomyoblasts. After the decellularization process the DNA content decreased from 758 (native) to 33 ng/mg dry tissue and the histological and SEM analysis showed absence of cells and nuclei in the tissue, demonstrating the success of this process, reducing chances to initiate a pro-inflammatory response that would influence negatively on tissue remodeling [3,4]. The histochemistry and collagen content analyses demonstrated preservation of the collagen in the scaffold after decellularization process. This preservation of collagen was critical for gel formation, allowing it to form a nanofibrous scaffold, seen by scanning electron microscopy. Collagen quantification also demonstrated that the addition of PRP to the hydrogel decreased the collagen content, as PRP was added vol/vol. After six days encapsulation into pure hydrogel, the MTT analysis showed that H9c2 cardiomyoblast had no increase in metabolic activity and proliferation compared to control group. On the other hand, this analysis evidenced that cell encapsulation in PRP-enriched hydrogels resulted in significant increases. Furthermore, the 20% group exhibited higher metabolic activity than the group encapsulated at 10%. The encapsulation also induced cell differentiation to cardiomyocyte-like after three days. Differentiated cells were observed in greater numbers in pure hydrogel group, followed by hydrogel+10%PRP then, hydrogel+20%PRP. In pure PRP group, no differentiated cells were seen. The results obtained in this work indicate that the injection of platelet-rich plasma enriched cardiac extracellular matrix hydrogel could assist cardiac tissue repair, once cardiac myoblasts would receive more stimuli for increased metabolic activity, proliferation, and differentiation. Furthermore, this biomaterial can be used for 3D bioprinting, as it is enriched with active biomolecules, which stimulate cell proliferation within the scaffold.

References

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