A study on hemocompatibility and *in-vitro* biodegradation properties of acellular caprine pericardium for tissue-engineering applications

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Abstract

Introduction: The acellular xenogeneic pericardium is a widely used biomaterial in the clinical application to design and fabricate tissue-engineered prostheses [1]. Hemocompatibility is the critical parameter for designing xenogeneic extracellular matrix in blood-contact biomaterial applications. This study focuses on the hemocompatibility and *in-vitro* biodegradation properties of the acellular caprine pericardium. Hemolysis, plasma clotting activity, and platelet activation assays were performed to understand the interaction of acellular pericardium when contact with whole blood. *In-vitro* enzyme biodegradability tests were performed on acellular pericardium with collagenase and trypsin enzymes to determine the degradation rate.

Methods: Caprine pericardium obtained from the local abattoir and fat deposits were removed, and the tissue specimen was subjected to decellularization. Briefly, the combination of sodium dodecyl sulfate and Triton X-100 was engaged to decellularize the caprine pericardium. Acellular matrix was incubated with anti-coagulated blood to determine the hemolysis rate. Isolated platelet-poor plasma was incubated with an acellular matrix, and then $CaCl_2$ was added to initiate the plasma recalcification time. Platelet-rich plasma is obtained from the whole blood and incubated with an acellular matrix to visualize platelet aggregates on the surface of the matrix. The acellular matrix's weight loss was determined by immersing in a 20 U/mL collagenase solution at different time intervals, and the weight loss of the samples was measured at regular time intervals. Similarly, 20 U/mL trypsin enzymes were used to determine the acellular matrix's weight loss.

Results & discussion: The intact acellular matrix was obtained after treatment with the combination of sodium dodecyl sulfate and Triton X-100. The absence of cell nuclei was confirmed through histological examination. The hemolysis rate of 3.7% for the acellular matrix pericardium met the clinically acceptable implant limit. A decrease in the plasma recalcification time shows that the acellular matrix regulates intrinsic and extrinsic pathways [2]. Platelet-rich plasma incubated on the acellular matrix surface shows no platelets' adhesion confirmed through SEM imaging. Due to porosity in the acellular matrix, the collagenase enzyme degrades rapidly within 12 hrs time interval. In the trypsin enzyme, the acellular matrix degrades continuously until 32 hrs. The presence of hidden cleavage sites of collagen and low solubility of the collagen fibrils causes a decrease in the activity [3].

References

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Introduction

- Pericardium derived from xenogeneic source is widely used to design and fabricate tissue-engineered bioprosthesis
- Hemocompatibility and in-vitro biodegradation is important aspect for the xenogeneic tissue when the material is designed for use in the vascular system where it is in direct contact with blood

Methods

- The combination of sodium dodecyl sulfate and Triton X-100 was engaged to decellularize the caprine pericardium. Then, the acellular matrix was incubated with anti-coagulated blood to determine the hemolysis rate
- Isolated platelet-poor plasma was incubated with acellular pericardium, and then $CaCl_2$ was added to initiate the plasma recalcification time
- Collagenase and trypsin enzymes were incubated with an acellular matrix at different time intervals to determine the degradation percentage

Highlights of the study

- **Decellularization pericardium shows better hemocompatibility and physicochemical properties compared to native tissue**
- **Caprine pericardium has a potential to be a biomaterial for tissue engineering applications**

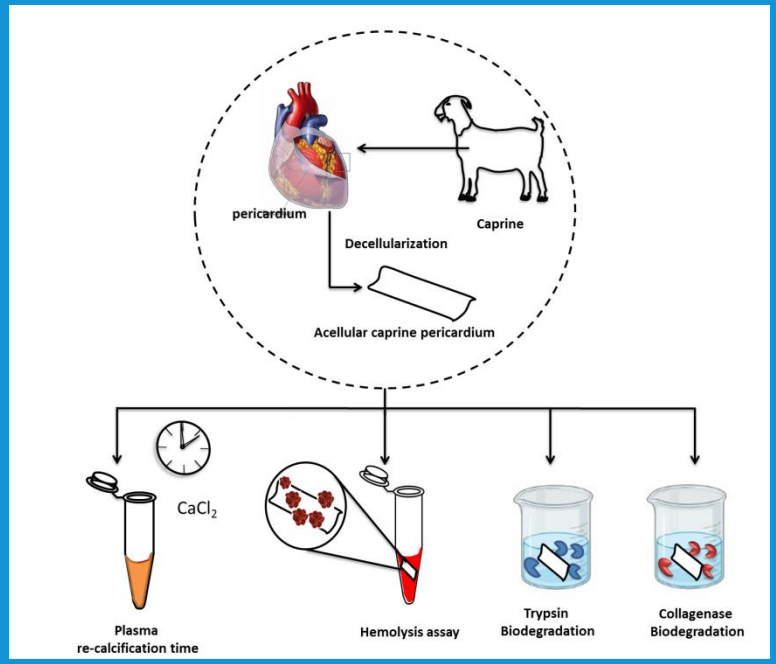

Figure 1: Graphical representation of the hemocompatibility and in-vitro biodegradation study on acellular caprine pericardium

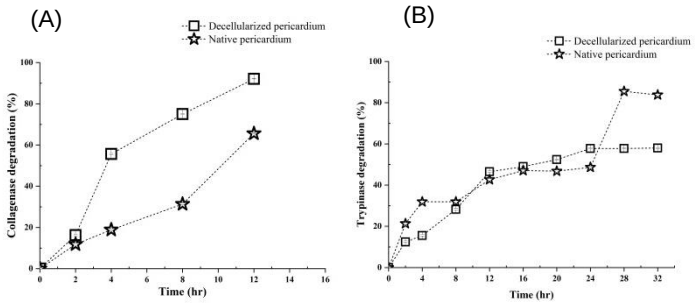

Figure 2: Collagenase and trypsin enzyme degradation study of the acellularized and native pericardium **(A)** Collagenase degradation, **(B)** Trypsin degradation

Results & Discussion

- The intact acellular caprine pericardium and absence of cell nucleus were confirmed through histological examination
- The hemolysis rate of 3.7% for the acellular pericardium met the clinically acceptable implant limit
- A decrease in the plasma recalcification time shows that the acellular pericardium regulates intrinsic and extrinsic pathways
- Due to porosity in the acellular pericardium, the collagenase enzyme degrades rapidly within 12 hr time interval