Investigation of pericardial decellularization with herbal agent for use in skin tissue engineering

Leila Rezakhani[1,2]
1.Fertility and Infertility Research Center, Health Technology Institute, Kermanshah University of Medical Sciences, Kermanshah, Iran
2. Department of Tissue Engineering, School of Medicine, Kermanshah University of Medical Sciences, Kermanshah, Iran

Kermanshah University of Medical Sciences

Derived from the design no 4040688

Introduction

Tissue engineering can be used to create scaffolds utilizing various cells or autograft, allograft, xenograft, or artificial Sources. There are many instances of organ and tissue loss worldwide because of disease, aging, trauma, and accidents. Donors and recipients of organ and tissue donation also face Challenges. Therefore, the utilization of extracellular matrix (ECM) in conjunction with tissue engineering, in a novel method known as decellularization, has been able to play a beneficial role in clinical applications. Biological scaffolds can also be decellularized using herbal agents, which have emerged as a promising protocol for replacing defective tissues. Residual cytotoxicity is one of the major disadvantages of non-herbal biological detergents. Numerous applications in tissue engineering and regenerative medicine have made successful use of tissue and organ decellularization. Because tissues and organs are so diverse, decellularization techniques and processes differ greatly. The origin and source of the tissue, its physical and chemical characteristics, and the enzymatic techniques employed all affect how well and efficiently cells can be extracted from it.

Methods

The 2-year-old male sheep pericardium of the Sanjabi breed was collected from the Kermanshah animal slaughterhouse in Iran. The adipose tissue was detached, and the samples were located in a bottle containing phosphate-bufferPericardial tissues were decellularized with different concentrations of ACP (5, 7.5% and 10%) and SDS (1%), as well as the combination of ACP + SDS. Tissue staining, biocompatibility (MTT), hemolysis, blood clotting index (BCI), scanning electron microscopy (SEM), ATR-FTIR spectroscopy, mechanical testing, contact angle, and antibacterial activity were performed.

Results

Complete cell removal was observed in the ACP + SDS combination groups, while the ECM structure was preserved. Biocompatibility was more than 90% in all groups. ACP-based scaffolds had less hemolysis, a more favorable coagulation index, preserved protein structure, higher porosity, and higher hydrophilicity. Although the mechanical properties were slightly reduced, they remained acceptable. The 10% ACP + 0.1% SDS group reported the highest antibacterial effect.

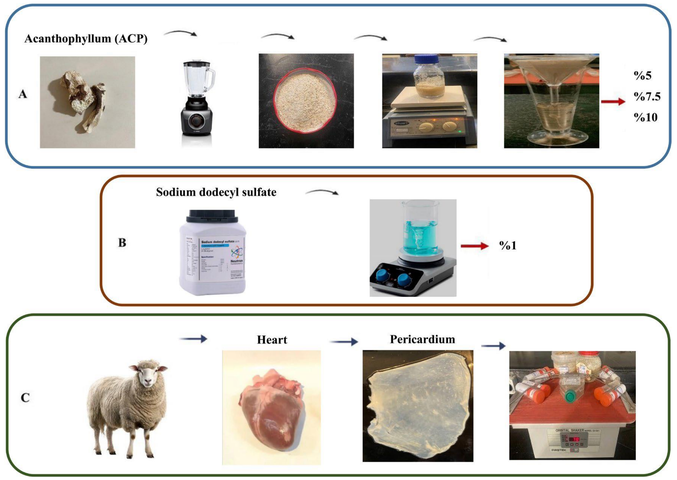

Figure 1. Decellularization Technique

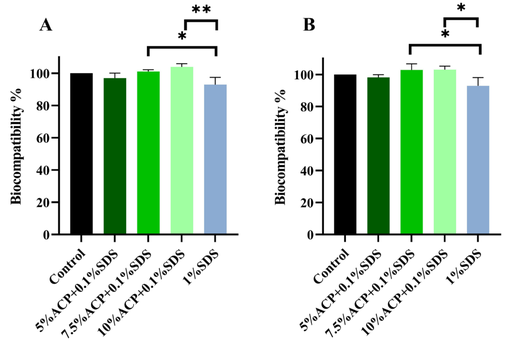

Figure 2. MTT assay, (A) 48 h, (B) 72 h, all experimental groups reported cell viability above 90%. No group showed significant differences compared to the control group. * ($p \leq 0.01$) and ** ($p \leq 0.002$), n = 3.

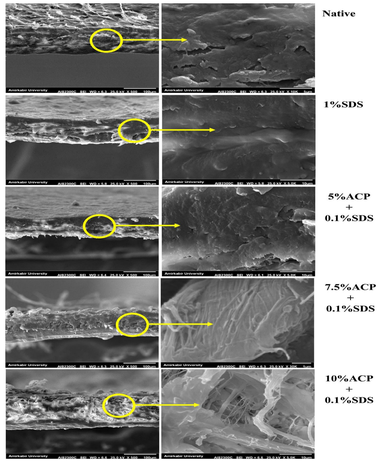

Figure 3. SEM images, in magnifications of X500, 5.0 K for native and decellularized pericardia

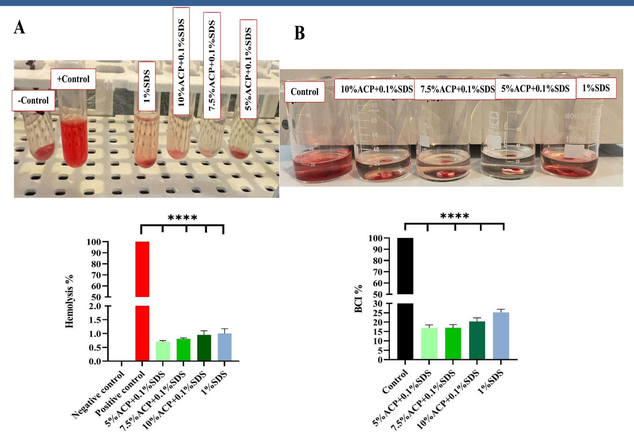

Figure 4(A) Hemolysis test, (B) BCI, **** ($p \leq 0.0001$), n = 3

Conclusions

The present study showed that Acanthophelium extract (ACP), especially in combination with low concentrations of SDS, can be used as an effective alternative to common chemical agents in the decellularization of tissues such as the pericardium. This decellularization agent was able to preserve the structure and mechanical properties of the extracellular matrix well, and at the same time did not reduce the biocompatibility. It also did not cause hemolysis. The scaffold obtained from the combination of 10% ACP and 0.1% SDS showed superior performance to other groups and had significant effectiveness against resistant bacteria, especially Acinetobacter baumannii and Staphylococcus aureus. These results indicate that the use of a combination of chemical and plant decellularization agents is a promising option for obtaining a suitable extracellular matrix for skin repair. It is recommended that further studies in animal models be conducted to investigate clinical efficacy.