Comparison of Collagen Matrices for an Advanced Transplantation Product aimed at Reduction of Complications after Pressure Injury Surgery

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Introduction

Pressure injuries (PI) are chronic wounds caused by increased pressure over a prolonged period in areas over bony prominences. PI are one of the most frequently occurring complications during hospitalization. The treatment of PI is very care intensive with long hospital stays and high complication rates. Recurrence of lesions remains a major problem in the long term.

A promising future direction to reduce complications and recurrences is the combination of tissue engineering techniques and regenerative medicine to create an implantable product augmenting the subcutaneous tissue after surgery. Previous studies suggest that stromal vascular fraction and autologous adipose derived stem cells (hADSC) may accelerate and improve wound healing. Furthermore, the combination of hADSC with bioactive autologous platelet rich plasma (PRP) could additionally enhance the healing process.

The aim of this study was to compare *in vitro* three different collagen scaffolds populated with hADSC or autologous fibroblasts from healthy dermis (hF) and from PI sites (hPIF). Other objectives were to test various combinations with human umbilical vein endothelial cells (HUVEC) and bioactive autologous platelet-rich plasma (PRP) to improve vascularization and adipogenesis of the tissue engineered constructs.

Materials and Methods

A number of monolayer (2D) and scaffold based (3D) cell culture experiments were conducted. As collagen scaffolds, BIOPAD (Euroresearch), Fibro-Gide and Mucograft (Geistlich) were trimmed to cubes of 3x3x3 mm and seeded with a combination of hADSC, hF, hPIF and HUVEC. The monolayers and cell cube constructs were incubated up to 18 days with different media and supplements like PRP in order to induce hADSC adipogenic differentiation and vascularization.

Outcome measures included cell proliferation, lipid quantification, RNA quantification and the expression of adipogenic and endothelial genes, evaluated via qRT-PCR, and immunohistochemical staining.

Results

Comparing the three collagen scaffolds, the Mucograft was excluded early on since it had the lowest cell survival and occasionally separated into its two layers which complicated the handling. The BIOPAD had a higher cell survival for HUVEC, while the Fibro-Gide had a highest cell survival with all other cells and cell combinations (see Fig. 1)

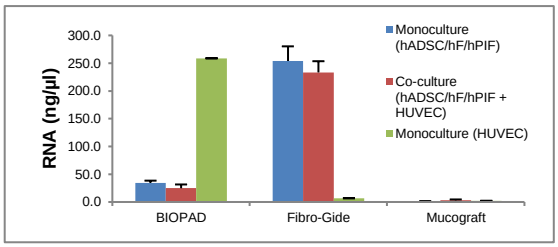

Fig. 1 RNA quantification in BIOPAD, Fibro-Gide and Mucograft cell cube constructs after 18 d incubation

For adipogenesis, the most promising results were obtained when hADSC + HUVEC were co-cultured into the BIOPAD or Fibro-Gide scaffolds. Pre-differentiation of hADSC before seeding them to the cell-cube constructs or adding HUVEC later on did not show any advantages.

Regarding angiogenesis of HUVEC in co-culture, the best results were also obtained with Fibro-Gide scaffold. There, HUVEC in co-culture with hPIF and hF had the highest endothelial gene expression. Additionally the best cell ratio of HUVEC to hADSC/hF/hPIF was 20-40% HUVEC. There was no observable difference between 4 and 9 days culture time. Surprisingly, added PRP did not improve angiogenesis compared to the control samples.

Discussion

According to this study, the most promising collagen scaffold over all is the Fibro-Gide, since it had the best cell survival, the most homogeneous cell distribution throughout the collagen cubes and the adipogenic differentiation worked well, especially when hADSC were in co-culture with HUVEC. Therefore, HUVEC ratio of around 30% and culture time of 4 days is advisable. Contrasting to the literature, PRP did not enhance angiogenesis in this study.

References

Smith, O.J., G. Jell, and A. Mosahebi, The use of fat grafting and platelet-rich plasma for wound healing: A review of the current evidence. International Wound Journal 16(1): 275-285, 2019

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