Strong human tissue-engineered blood vessels : cultured in weeks instead of months

Strong Human Tissue-Engineered Blood Vessels: Cultured in Weeks instead of Months

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Introduction

Vascular tissue engineering represents a promising approach for the development of living small diameter blood vessels that can be used for replacement therapy, including coronary artery bypass grafting (CABG). So far, the culture of strong human tissue-engineered (TE) blood vessels required long culture times, up to several months, whether or not combined with telomerase gene therapy. In the present study we describe the culture of strong, living, human TE blood vessels in 28 days.

Materials&Methods

Blood vessel constructs (n=8) were fabricated from P4HB coated PGA (Fig. 1). Myofibroblasts were harvested from the saphenous vein of CABG patients and the tubular constructs were seeded using fibrin as a cell carrier.

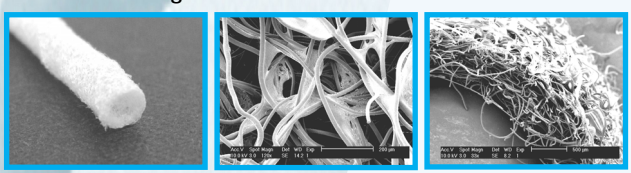

Figure 1: *Blood vessel construct of P4HB coated PGA*

The TE blood vessels were cultured in a bioreactor for 4 weeks. To improve the tissue formation, tubular constructs were constrained in axial direction and dynamically strained in circumferential direction (Fig. 2). The properties of the engineered blood vessels were compared to those of native CABG grafts, i.e., the left internal mammary artery (LIMA, n=3) and the saphenous vein (SV, n=5).

Results

Living TE blood vessels after 4 weeks of in-vitro culture (Fig. 3) showed dense tissue formation. Uniaxial tensile tests showed that the vessels were significantly stronger and stiffer in axial than in circumferential direction (Fig.4).

The burst pressure of the TE vessels was 906±123mmHg (n=4). The experiments showed that the strain at 100mmHg was approximately 10%. The compliance of the vessels was in the order of 0.03%/mmHg.

Figure 2: *bioreactor set-up*

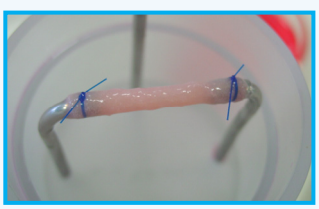

Figure 3*: Living TE blood vessel in bioreactor after 4 weeks of in-vitro culture*

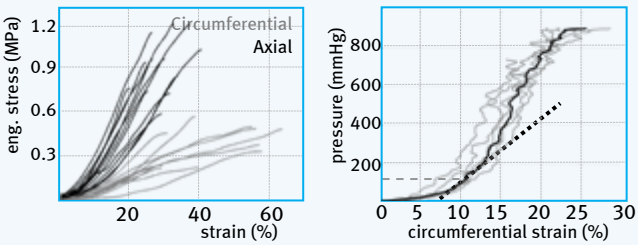

Figure 4*: Uniaxial tensile tests (left) and burst pressure measurement (right) of TE vessels*

Histological examination revealed significant amounts of collagen in the TE blood vessel, although less, and not as organized as in the native vessels. (Fig. 5)

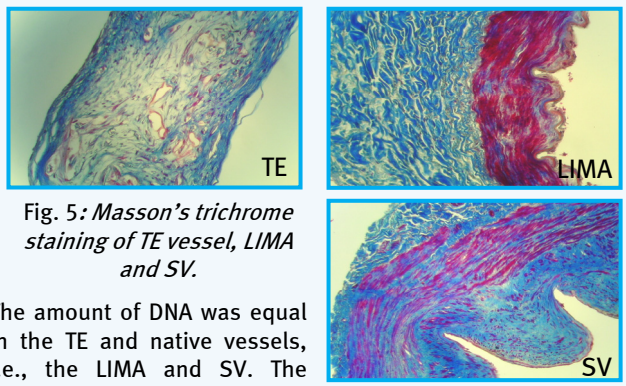

Fig. 5*: Masson's trichrome staining of TE vessel, LIMA and SV.*

The amount of DNA was equal in the TE and native vessels, i.e., the LIMA and SV. The amount of glycosaminoglycans was higher in the TE constructs than in the native vessels, whereas the amount of hydroxyproline, as a measure for collagen, was 50% of native (Fig. 6).

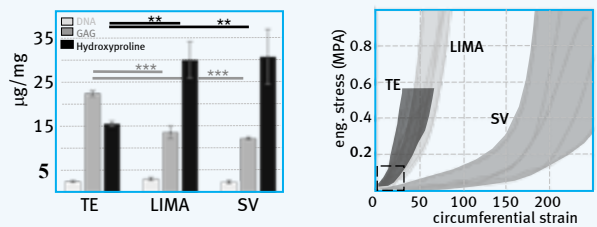

Figure 6*: DNA, GAG and hydroxyproline content of TE and native vessels (left). Circ. tensile tests at low stress values (right).*

Tensile tests showed that the mechanical behavior of the TE blood vessels resembled that of native arteries in the physiologically relevant range (Fig. 6, dashed box).

Conclusions

In this study we present the strongest human TE blood vessels in a 28d-culture period, in which the scaffold no longer contributed to the mechanical properties. Mechanical behavior of the TE vessels resembled that of native arteries in the physiologically relevant range.