

7 SUMMARY

Background: One approach to tissue engineering has been the development of *in vitro* conditions for the fabrication of functional cardiovascular structures intended for implantation. In the current experiment, we developed a pulsatile flow system that provides biochemical and biomechanical signals in order to regulate autologous patch-tissue development *in vitro*.

Methods: We constructed a biodegradable and biocompatible patch scaffold from porous poly-4-hydroxy-butyrates (P4HB; pore size: 80-150 micron, thickness: 0.5 mm, diameter: 45 mm; Tepha Inc., USA). The porous patch scaffold was seeded with pediatric aortic cells. The cell-seeded patch constructs were placed in a self-developed bioreactor, providing pulsatile flow and biomechanical stress for 7 days to observe potential tissue formation under dynamic cell culture conditions. As a control, porous patch scaffolds were seeded with pediatric aortic cells using the same experimental protocol without conditioning in our bioreactor system. Following maturation *in vitro*, analysis of the tissue engineered constructs included biochemical, morphological and immunohistochemical examination.

Results: Macroscopically, all tissue engineered patch constructs were covered by tissue. After conditioning in the pulsatile flow bioreactor, the cells were mostly viable, grew into the pores and formed tissue on the porous patch construct. Electron microscopy showed smooth confluent smooth surfaces over the follow-up period. Additionally, we demonstrated the capacity to generate collagen and elastin under *in vitro* pulsatile flow conditions in our biochemical examination. Immunohistochemical examination stained positive for alpha-smooth muscle actin, collagen type 1 and fibronectin. There was minor tissue formation in the non-conditioned control samples.

Conclusions: Porous P4HB may be used to fabricate a porous and biodegradable patch scaffold. The cells attached themselves to the polymeric scaffold, and extracellular matrix formation was induced under controlled biomechanical and biodynamic stimuli in a self-developed pulsatile bioreactor system.