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Novel collagen and ceramic based 3D biomaterials for Tissue Engineering applications

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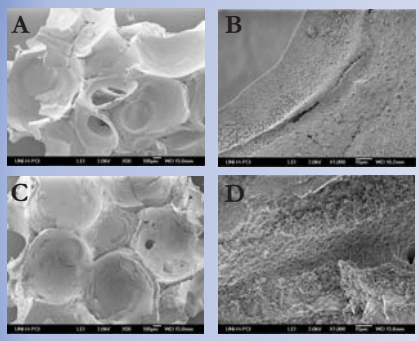
Introduction

The principle of Tissue Engineering is to build artificial replacements by means of vital components. In order to achieve a functional graft, applicable cells were seeded onto three-dimensional scaffolds and expanded *in vitro*. The commonly used cell culture techniques generate cell layers, but it is not possible to create a three dimensional, functional multilayer cell structure on the surface of a cell culture dish. Therefore, three dimensional scaffolds are necessary, which provide a specific environment and architecture for the formation of the tissue.
Numerous materials have already been tested for their applicability, and both ceramics and natural and synthetic polymers are the most promising materials for Tissue Engineering. In this study, we characterized collagen- and ceramic based scaffold materials with regard to their biomechanical properties. Furthermore, the materials were tested in cell culture under static and dynamic conditions.

Biomechanical testing



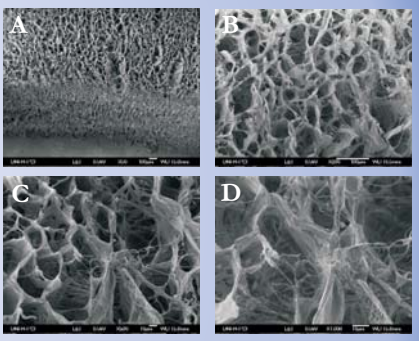
SEM pictures of Sponceram®, a macroporous ZrO_2 ceramic. A,B: Sponceram®; C,D: hydroxyapatite coated Sponceram®(Sponceram®/HA)
Magnification: 50 times (A, C) and 1000 times (B,D)

Material	Permeability [D]
Sponceram® 30/90	2726.838 ± 371.106
Sponceram® 30/90 HA	651.023 ± 6.396
Sponceram® 30/90 Ti	666.127 ± 8.499
Sponceram® 30/145	394.832 ± 3.183

Material	Max. Deformation at 0,25 kN [%]	Max. Tension [MPa]
Sponceram® 30/90	7.660 ± 2.108	0.677 ± 0.181
Sponceram® 30/90 HA	7.168 ± 1.331	1.085 ± 0.125
Sponceram® 30/90 Ti	5.6360 ± 1.094	0.9205 ± 0.0718
Sponceram® 30/145	7.645 ± 0.829	1.0471 ± 0,049

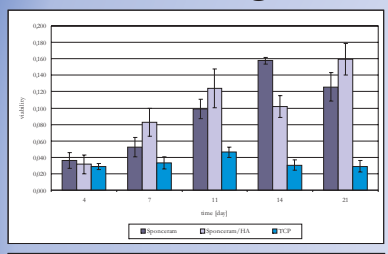
Material	Deformation at 4.5 N [%]	Tension at 25 % Deformation [kPa]
Matristypt®	88.434 ± 0.111	0.812 ± 0.061
Matriderm®	85.310 ± 0.260	1.071 ± 0.113
Decellularized skin	54.668 ± 0.454	5.664 ± 0.561

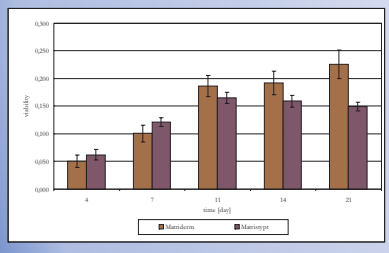
The materials were characterized with regard to their permeability, compressive deformation and tension. The Sponceram based materials differed in their permeability but showed similar deformation and tension by mechanical load. The collagen based materials showed comparable deformation and tension.



SEM pictures of Matriderm®, a collagen-elastin meshwork
Magnifications: 50 times (A), 200 times (B), 500 times (C), 1000 times (D)

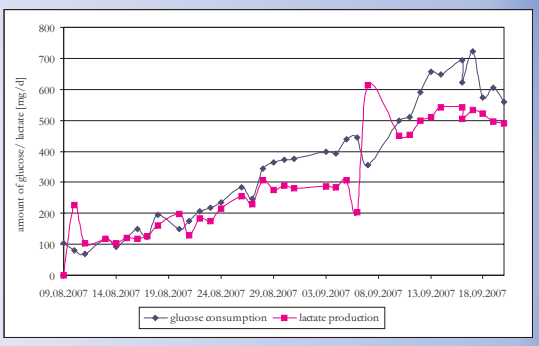
Cell culture testing

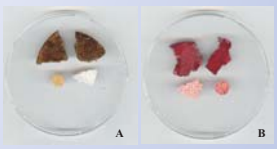


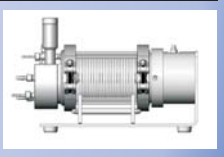


For the static cell culture testing, human mesenchymal stem cells derived from fat tissue were seeded onto ceramic [Sponceram®, Sponceram®/HA, Tricalciumphosphate (TCP)] and collagen materials [Matriderm®, Matristypt®]. 4, 7, 11, 14 and 21 days after cell seeding, the MTT assay was performed to determine the viablility of the cultivated cells. The results demonstrate that the viability of the cells on the Sponceram® matrices is increased compared to TCP. The cells cultivated on the collagen-elastin material (Matriderm®) show an increased viability compared to the pure collagen matrix (Matristypt®).

For the dynamic cultivation, human mesenchymal stem cells derived from fat tissue were seeded onto Sponceram® and Sponceram®/HA discs and were cultivated in a rotating bed system (Z®RP) for 47 days using osteogenic differentiation medium (containing dexamethason, β-glycerolphosphate, ascorbic acid).
The continuously increasing consumption of glucose and production of lactate show a good cell proliferation.
Von Kossa (A) and Alizarin red staining (B) show calcification on the Sponceram matrices.







Discussion and Outlook

The performed experiment showed, that the testet Collagen and Sponceram® materials are appropiate for the cultivation of mesenchymal stem cells. The cells which were cultured on Sponceram® in osteogenic differentiation medium under dynamic conditions have deposited extracellular matrix after 6 weeks.
In the future, the rotating bed reactor (Z®RP system) should be used to test different functionalized sponceram materials with regard to osteogenic differentiation.

Acknowledgement

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