Three-Dimensional Controlled Fluid Environment Promotes Osteogenic Differentiation of Mesenchymal Stem Cells

Birgit Weyand¹, Cornelia Kasper², Meir Israelowitz³, Christoph Gilles³, Herbert P. von Schroeder³, Kerstin Reimers¹, Peter M. Vogt¹
weyand.birgit@mh-hannover.de

¹Department of Plastic, Hand and Reconstructive Surgery, Hannover Medical School, Hannover, Lower Saxony, Germany, ²Institute of Technical Chemistry, Leibniz University, Hannover, Lower Saxony, Germany, ³Biomimetics Technologies Inc, Toronto, Ontario, Canada, ⁴Department of Surgery, University Hand Program and Bone Lab, University of Toronto, Toronto, Ontario, Canada

Introduction
Mechanical stressors like pressure, gravity, stretch or fluid shear stresses can influence cell morphology, migration, proliferation and differentiation. We have recently developed a laminar flow bioreactor for cell cultivation within a porous scaffold which allows flow constancy during cellular growth within the pores¹,². The objective of this work was to study the effect of controlled flow shear stresses on human adipose mesenchymal stem cells within a porous matrix cultured in the laminar flow bioreactor.

Materials and Methods
Primary human adipose mesenchymal stem (haMSCs) cells were obtained from abdominal fat tissue of patients undergoing abdominoplasty by collagenase digestion. Cells were expanded and cultured in stem cell media without addition of differentiating agents such as dexamethasone or specific growth factors. Stem cells were characterized by flow cytometry and by their capacity to differentiate into the adipogenic, osteogenic and chondrogenic pathway. Stem cell-seeded macroporous ceramic scaffolds and microporous collagen matrices were cultured inside a laminar flow bioreactor for up to 3 months and analyzed by surface microscopy, grinding sectioning, extracellular matrix staining and electronmicroscopy.

Results
Cell-seeded porous matrices cultured in the laminar flow reactor demonstrated homogenous cell distribution and cell growth within pores. Life-dead-staining assays verified high cell viability inside the matrices after a five week culture period within the bioreactor. The rate of flow perfusion with specific shear stresses affects cell morphology and amount of extracellular matrix mineral deposition compared to static controls as revealed by raster electron microscopy.

Discussion and Conclusions
Controlled flow shear stress can stimulate osteogenic differentiation and extracellular matrix deposition of human adipose mesenchymal stem cells without addition of external stimuli such as dexamethasone to the culture medium. The amount of extracellular matrix deposition depends on flow rates and shear stresses. Further studies are needed to understand the underlying mechanisms of shear stress-induced osteogenic differentiation of mesenchymal stem cells within a three-dimensional fluid environment.

References

Acknowledgments
This work was supported by an internal start-up grant from Hannover Medical School.

Disclosures
The authors state no conflict of interest.