TISSUE AND ORGAN MECHANOBIOLOGY

Benjamin Gantenbein, Head of Research Group
Email: benjamin.gantenbein@istb.unibe.ch
Phone: +41 31 631 59 51


Benjamin Gantenbein


Ezgi Bakirci


Martina Caliò


Emina Džafo


Daniela Frauchiger


Rahel May


Selina Steiner


Simon Wuest


Xingshuo Zhang

Clinical Partners Bern:

Dr med Christoph Albers, Department of Orthopaedics, Spine, Insel Hospital, University of Bern
Prof Dr med Lorin Benneker, Department of Orthopaedics, Head of Spine, Insel Hospital, University of Bern
PD Dr med Frank Klenke, Department of Orthopaedics, Knee Team, Insel Hospital, University of Bern
Dr med Sven Hoppe, Department of Orthopaedics, Spine, Insel Hospital, University of Bern
Prof Dr Klaus Siebenrock, Department of Orthopaedics, Insel Hospital, University of Bern
Prof Dr med Moritz Tannast, Department of Orthopaedics, Hip Surgery, Insel Hospital, University of Bern
Prof Dr Paul Heini, Spine Surgeon, Orthopaedics, Sonnenhof Clinic

Research Profile

The Tissue & Organ Mechanobiology (TOM) Group of the Institute for Surgical Technology and Biomechanics (ISTB), University of Bern, conducts translational research in the intersection of tissue engineering, biology and applied clinical research. The group's primary aim is to understand the cellular response onto biomechanical stimuli and how cellular communities are affected *in situ* using 3D tissue and organ culture models. Our research can be divided into two main foci: On the one hand the group investigates causes of low back pain due to intervertebral disc (IVD) degeneration and on the other hand the group focuses on the human knee where they aim to identify cell-based solutions for the non-healing or delayed ruptures of the anterior cruciate ligament (ACL). The common focus of the TOM group is to advance *in vitro* organ culture models, which match closely the human situation and where regenerative therapy strategies, such as novel biomaterials and cells, can be tested in a most authentic *in vitro* set-up.

Low Back Pain and Intervertebral Disc Degeneration and Regeneration

The TOM group conducts research in two main directions: i) IVD research in the area of regeneration using biomaterials and stem cells[1-4] and ii) in the area of non-successful spinal fusion and possible involvement of pseudo-arthrosis[5]. For the first research area we use a combination of 3D tissue and organ culture approaches. The research of the second focus is the understanding of the balance between BMP agony and antagony. Besides the investigation of the exogenous stimulation of BMP antagonists on mesenchymal stem cells (MSC) and osteoblasts, the main focus lies on the observation of the interaction between IVD cells and osteoblast, by performing co-cultures[1].

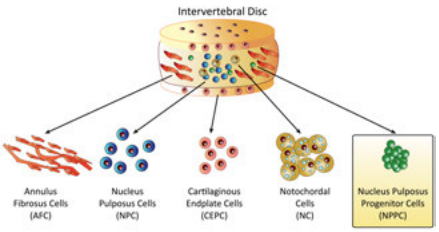

Figure 1. Image illustrating the four "classical" cell populations previously characterized in the intervertebral disc. In yellow on the right are the newly detected Tie2+ nucleus pulposus progenitor cells (NPPC).

In a Gebert Rüf financed project a fiber-reinforced hydrogel was tested in a physiologically clinically relevant organ culture model by cross-linking the fibrin mesh with genipin[1]. Therefore, a healthy control, an injured IVD (2 mm biopsy punch) and the repaired IVD were tested, and histology was performed to visualize the injury and integration of the novel silk and fibrin hydrogel. These results were recently reported in the Journal of Functionalized Biomaterials[1] and in the European Spine Journal[3].

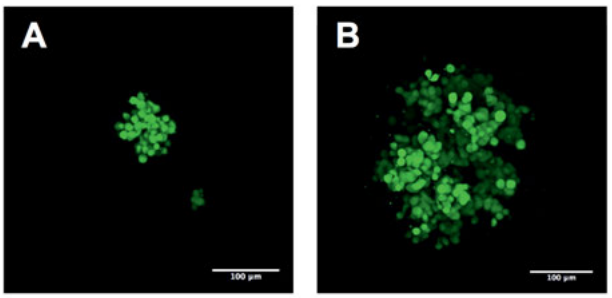

Figure 2. Confocal Laser Scanning Microscopy of A) nucleus pulposus progenitor cells (NPPC) and B) nucleus pulposus cells (NPC) after seven days of colony unit forming assay in a viscous medium. NPPC do result in more dense and spherical colonies whereas NPC form more lose and wider spread colonies. Cells were stained with a live dye in green. Scale bar = 100 µm.

Recently, autochthonous progenitor cells were detected in the human IVD, which could lead the path to cell therapy (Figure 1). Here, we concentrated on the most suitable isolation protocols to "fish" nucleus pulpous progenitor cells (NPPC) from the total population of cells in the bovine coccygeal disc. We also focused on their multipotency capacity and their application for IVD repair (Figure 2). Future research is to understand how these cells can be best isolated and whether these cells can be maintained *in vitro* to regenerate the IVD[4]. Furthermore, it would be highly desirable to investigate how induced multipotent stem cells (iPSC) could be used for IVD repair. This is the main aim in an upcoming Horizon 2020 Project named "iPSpine" starting in 2019 for three years in collaboration with internationally well-known scientists and experts in the field of engineering, biomaterials and biomechanics.

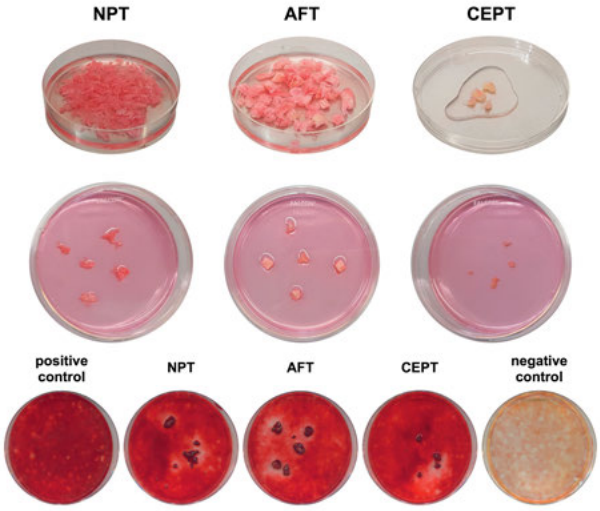

Figure 3. Human mesenchymal stromal cells (hMSC) were seeded in 100 mm Petri dishes and cultured until they reached 90% of confluency. Human intervertebral disc (IVD) explants (tissue of the nucleus pulposus (NPT), annulus fibrosus (AFT) or cartilaginous endplate (CEPT), 2-5 mm3) were cultured in direct contact with the hMSC in osteogenic medium (lacking bone morphogenetic protein 2). Top row: preparation of tissue. Middle row: Contribution of human NPT, AFT and CEPT on the top of hMSC monolayer. Bottom row: Alizarin red staining of direct culture after stimulation of 21 days with osteogenic medium (except negative control) and co-cultured with NPT, AFT and CEPT. Proof-of-concept of inhibitory effects (N = 1).

Biological Repair of the ruptured Anterior Cruciate Ligament

In Switzerland, the incidence of ACL ruptures is estimated at 32 per 100,000 in the general population and even more than double in the sports community. The current gold standard for ACL repair is reconstruction using an autograft, however, this approach has shown some limitations. Here, cell-based approaches using collagen patches or the application of platelet-derived plasma (PRP) are of interest for the clinical application.

Reducing the Senescence in Mesenchymal Stem Cells

Stem cell therapy faces the problem of the necessity to rely on fetal bovine serum (FBS) for cell expansion, which proved to have major disadvantages for application in the clinics. Additionally, MSC undergo senescence during expansion *in vitro*, which impairs their therapeutic potential. Here, alternate serum-free media formulations were investigated in terms of cell proliferation and differentiation potential, which could make their way to a GMP-compliant solution.

Selected Publications

1. Frauchiger DA, May RD, Bakirci E, Tekari A, Chan SCW, Wöltje M, Benneker LM, Gantenbein B, Frauchiger D, May R, Chan S, Benneker L (2018) Genipin-Enhanced Fibrin Hydrogel and Novel Silk for Intervertebral Disc Repair in a Loaded Bovine Organ Culture Model. J Funct Biomater 9(3):40. doi: 10.3390/jfb9030040
2. Frauchiger DA, Heeb SR, May RD, Wöltje M, Benneker LM, Gantenbein B. Differentiation of MSC and annulus fibrosus cells on genetically engineered silk fleece-membrane-composites enriched for GDF-6 or TGF-β3. J Orthop Res 2018; 36:1324-1333 DOI: 10.1002/jor.23778.
3. Frauchiger DA, Chan SCW, Benneker LM, Gantenbein B (2018) Intervertebral disc damage models in organ culture: a comparison of annulus fibrosus cross-incision versus punch model under complex loading. *Eur Spine J*: doi: 10.1007/s00586-018-5638-5
4. Sakai D, Schol J, Bach FC, Tekari A, Sagawa N, Nakamura Y, Chan SCW, Nakai T, Creemers LB, Frauchiger DA, May RD, Grad S, Tryfonidou MA, Gantenbein B (2018) Successful fishing for nucleus pulposus progenitor cells of the intervertebral disc across species. *JOR Spine* 2018e: doi: 10.1002/jsp2.1018
5. May RD, Frauchiger DA, Albers CE, Benneker LM, Kohl S, Gantenbein B (2018) Inhibitory Effects of Human Primary Intervertebral Disc Cells on Human Primary Osteoblasts in a Co-Culture System. *Int J Mol Sci* 19(4): doi: 10.3390/ijms19041195
6. Wuest SL, Caliò M, Wernas T, Tanner S, Giger-Lange C, Wyss F, Ille F, Gantenbein B, Egli M. Influence of Mechanical Unloading on Articular Chondrocyte Dedifferentiation. *Int J Mol Sci* 2018; 19: [DOI: 10.3390/ijms19051289
7. Wuest SL, Gantenbein B, Ille F, Egli M. Electrophysiological experiments in microgravity: lessons learned and future challenges. *NPJ Microgravity* 2018; 4:7 DOI: 10.1038/s41526-018-0042-3

Selected Conference Contributions

1. Bakirci E, Hugi A, Ahmad S,Kohl S, Guenat O & Gatenbein B. Optimization of 3D Bioprinted Hydrogels with Primary Anterior Cruciate Ligament Cells for Tissue Engineering Applications. *International Conference on Biofabrication* 2018, 28-31 October, Wuerzburg Germany.
2. Caliò M, Wuest SL, Wernas T, Tanner S, Giger-Lange C, Wyss F, Ille F, Gantenbein B, Egli M. Influence of mechanical unloading on articular chondrocyte dedifferentiation, in eCM Online Periodical, 2018, *eCM Conference Abstracts, 2018 eCM XVIII: Cartilage & Disc: Repair and Regeneration, 25-28 June* 2018, Davos.
3. Frauchiger DA, Chan SCW, Benneker LM, Gantenbein B. Comparison of two annulus fibrosus injury models investigated in a loaded bovine intervertebral disc organ culture format, in eCM Online Periodical, 2018, eCM Conference Abstracts, 2018 eCM XVIII: Cartilage & Disc: Repair and Regeneration, 25-28 June 2018, Davos.
4. Frauchiger DA, May RD, Zhang X, Stoyanov J, Bertolo A, Benneker LM, Grad S, Tryfonidou MA, Gantenbein B. Comparing three cell isolation techniques for "fishing" angiopoetin-1 (Tie 2) positive progenitor cells from the nucleus pulposus, in eCM Online Periodical, 2018, *eCM Conference Abstracts, 2018 eCM XVIII: Cartilage & Disc: Repair and Regeneration, 25-28 June* 2018, Davos.
5. Gantenbein B. Invited Keynote: Repair of the Intervertebral Disc using Biomaterials and Progenitor Cells, in *Proceedings of Biospine Asia Pacific, 26-28 April* 2018, Seoul, South Korea.
6. May RD, Frauchiger DA, Benneker LM, Gantenbein B. Comparison of gene expression of discs from Diffuse Idiopathic Skeletal Hyperostosis (DISH) and trauma patient, in *Proceedings of Biospine Asia Pacific, 26-28 April, Poster Presentation* 2018, Seoul, South Korea.
7. May RD, Frauchiger DA, Benneker LM, Gantenbein B. Osteoinductive Stimulation of Intervertebral Disc Cells with Bone Morphogenetic Protein 2 or Osteogenic Medium, in *Proceedings of Biospine Asia Pacific, 26-28 April, oral Presentation 2018,* Seoul, South Korea.
8. Gantenbein B. Oral Presentation: Mechanical loading under compression and torsion of bovine coccygeal intervertebral discs, in *Proceedings of the 8th World Conference of Biomechanics, 8-12 July* 2018, Dublin.