7th KMM-VIN Industrial Workshop

Biomaterials: Key Technologies for Better Healthcare

Programme and Abstracts

Institute of Biomaterials
FAU Erlangen-Nuremberg

InnoPlanT/Medical Valley Centre

FORM Laboratory-UK Erlangen

European Virtual Institute on Knowledge-based Multifunctional Materials KMM-VIN AISBL

27th-28th September 2017
Erlangen, Germany

Biomaterials Erlangen

MEDICAL VALLEY
INNOPLANT.NET

FORM Forschungslabor Orthopädie für Radiostereoanalyse und Medizintechnik

FAU FRIEDRICH-ALEXANDER UNIVERSITÄT ERLANGEN-NÜRNBERG

KMM.VIN

About organizers

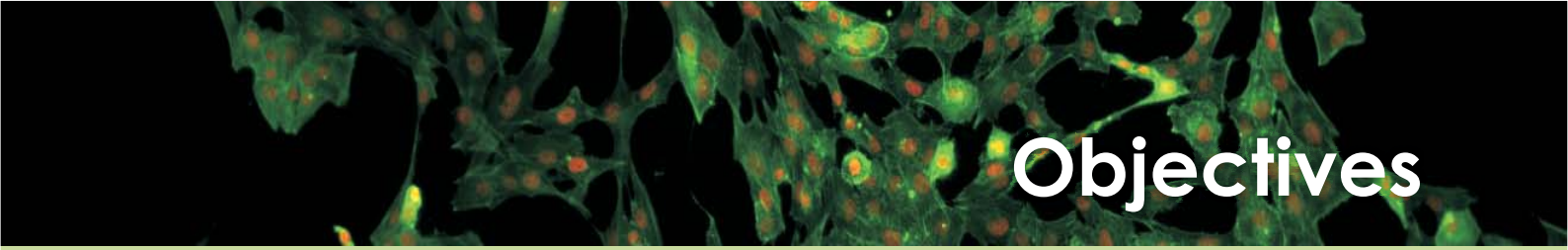
KMM-VIN AISBL is an international non-profit association that creates conditions for networking and conducting joint research on advanced materials.

It offers mobility programme for young researchers and customised courses and trainings with focus on materials for Transport, Energy and Biomedical sectors.

Institute of Biomaterials at the University of Erlangen-Nuremberg is an internationally recognised leader in the field of biomedical materials research, carrying out a wide range of interdisciplinary research projects on multifunctional bioactive materials for implant applications as well as for tissue engineering, biofabrication and drug delivery.

InnoPlanT.NET is a community of practice of the **Medical Valley EMN**. The network serves as a platform for researchers and companies developing implants and transplants. One of the main objectives of **InnoPlanT** is the initiation of research projects in the field. Regular network meetings are considered as a source of ideas to start new cooperations.

FORM Laboratory is an integral part of the Department of Orthopaedic Surgery of the University of Erlangen-Nuremberg. FORM focusses on interdisciplinary fundamental and clinical research projects on implants in the field of orthopedics and traumatology, mainly for quality control and improvement of medical devices



Objectives

The goal of this workshop is to bring together experts working in the broad area of biomedical materials both in industry and academia, covering topics related to design, fabrication, characterisation and applications of a wide variety of biomaterials.

Materials science and technological aspects, cell biology and in vivo evaluation of new biomaterials as well as analysis of the performance of established biomedical materials in clinical applications will be discussed. In addition, contributions are invited to cover advanced concepts of tissue engineering and regenerative medicine, including biofabrication approaches, controlled drug delivery systems and biomaterials models for in vitro screening and for cancer research and therapy. The workshop will include but will not be limited to the following topics:

- Biomaterials for bone and dental implants
- Biomaterials for cardiovascular applications
- Bone filling materials
- Scaffolds for hard and soft tissue engineering
- Bioactive and cell instructive materials
- Drug delivery carriers
- Biomedical coatings
- Hydrogels and biofabrication approaches
- 3D biomaterial models for cancer research
- Clinical performance of biomaterials
- Biosensors and theranostics

Venue

Medical Valley Center Erlangen, Hörsaal ZMPT (Room 01.020; 1st Floor)

Henkestrasse 91, 91052 Erlangen, Germany.

Workshop programme

27th September 2017

Registration and coffee (12:00-12:50)

Opening & Welcome (12:50-13:00) – Aldo R. Boccaccini (FAU), Michał Basista (KMM-VIN)

Keynote lectures I (13:00-15:00) – *Chair*: Aldo R. Boccaccini

- Functionalization of surfaces and applications in healthcare surroundings – **Frank Heidenau** (BioCer Entwicklungs-GmbH)
- Opportunities and challenges of 3D printing in healthcare applications - **Tim Van Cleynenbreugel** (3D Systems)
- Requirement for Ceramic Implants for Hip Joint Prostheses – **Thomas Oberbach** (Mathys Medical AG)
- Soluble Phosphate Glasses and Composites as Key Biomaterials – **David M. Healy** (IDP Services Ltd)

Coffee break (15:00-15:30)

Keynote lectures II (15:30-16:30) – *Chair:* Stefan Sesselmann (FAU)

- When cell meets textiles – towards biohybride implants - **Stefan Jockenhövel** (RWTH Aachen University)
- LIPUS – Low Intensity Pulsed Ultrasound for bone healing purposes: Only a gimmick or a mechanism-based bone regenerative approach? - **Jochen Salber** (University Hospital Knappschaftskrankenhaus Bochum)

Oral session I (16:30-18:00) *Chair*: Christian Hellmich (TU Vienna)

Surface modifications at the nanoscale for a better integration of implants to the hard and soft tissues and a reduced bacterial contamination – **S. Spriano**, F. Baino, C. Balagna, M. Cazzola, A. Cochis, M. Ferraris, S. Ferraris, M. Miola, C. Ramskogler, L. Rimondini, A. Varesano, E. Vernè, C. Vineis, F. Warchomicka

Site-directed Immobilization of BMP-2 onto collagen beads for bone regeneration – **C. Siverino**, J. Nickel, H. Walles

Electrospun multilayered scaffolds for interface tissue engineering applications – **L. Liverani**, A.R. Boccaccini

Influence of polymer film concentration on cytocompatibility and corrosion suppression of ZM21 magnesium alloy – **A. Witecka**, A. Yamamoto, W. Święszkowski, M. Basista

Hybrid materials for biomedical applications – **J.C. Almeida**, I.M.M. Salvado, M.H.V. Fernandes

Single cell analysis for studying tumor heterogeneity – L. Hoene, A. Schwab, B. Fabry, M. Théry, A.R. Boccaccini, **A. Leal-Egana**

Technical break (18:00-18:15)

27th September 2017 (continued)

Short oral presentations (18:15-19:15) – *Chair*: Aldo R. Boccaccini (FAU)

1. Exploiting piezoelectric polymeric platforms for neuronal tissue engineering – **M.H. Fernandes**, N. Barroca, A. Marote, A. Almeida, S.I. Vieira, O. da Cruz e Silva, P.M. Vilarinho

2. Personalised 3D-printed smart eyeglasses for physiological monitoring – **O. Amft**, R. Zhang, F. Wahl

3. Stem cell labeling with iron oxide nanoparticles in a 3D environment – **M. Steinke**, T. Kilian, C. Grüttner, S. Hackenberg, F. Fidler, K. Schütze, H. Walles

4. Wedge-shape compression of MSC-laden Col I gel for meniscus tissue engineering – **A. Kremer**, J. Reboredo, H. Walles

5. Development of bioactive bioglass scaffolds coated with iron-loaded hydroxyapatite nanocomposites as potential biomaterials for bone tissue repair – **M.L. Dittler**, M.C. Gonzalez, A.R. Boccaccini

6. Development and characterization of hybrid hydrogel based on alginate di-aldehyde, gelatin and silk fibroin for tissue engineering applications – **S. Reakasame**, A.R. Boccaccini

7. Functional polyelectrolyte coatings in prevention of medical devices from bacterial colonization – **A. Mzyk**, J. Straub, M. Riool, L. de Boer, R. Major, S.J. Zaat

8. Electrophoretic deposition of gentamicin loaded chitosan/gelatin/bioactive glass composite coatings on PEEK/bioactive glass layers: A comprehensive study on in-vitro-bioactivity and antibacterial effect – **M.A. Ur Rehman**, W.H. Goldmann, A.R. Boccaccini

9. Tribocorrosion response of different multilayer TaN coatings in biological environments – R. Bayón, **V. Sáenz de Viteri**, L. Mendizabal, J. Barriga, A. Igartua

10. Pure titanium with enhanced properties applied to newly developed dental implant – K. Sztwiertnia, J. Kawalko, **M. Bieda**, D.Wojtas, K. Wierzbanowski, M. Wronski, W. Pachla, M. Kulczyk

11. Development of a conductive, biomimetic, polymeric fiber mat for a bilayered cardiac patch – **L. Vogt**, F. Ruther

12. Cinnamon bark oil encapsulated poly (ε-caprolactone) nanofiber mats for wound healing – **I. Unalan**, A.R. Boccaccini

13. Synthesis, characterisation and applications of ordered mesoporous silver doped bioactive glass – **F.E. Ciraldo**, A.R. Boccaccini

14. Electrophoretic deposition of zein/bioactive glass composites – **L. Ramos-Rivera**, N. Meyer, L. Tortoreto, A.R. Boccaccini

15. Biodegradable zinc alloy with magnesium subjected to hydrostatic extrusion: evolution of microstructure, mechanical properties and corrosion behavior – **A. Jarzębska**, M. Bieda, Ł. Rogal, R. Chulist, J. Guśpiel, M. Strąg, K. Sztwiertania, W. Pachla, M. Kulczyk

16. Preparation and characterization of mesoporous calcium doped silica coated TiO_2 scaffolds and their drug releasing behavior – **A. Sengottuvelan**

17. Applications of low energy electron beam technology for sterilization and surface modification of medical products – **G. Gotzmann**

18. The effect of chemical composition on viscoelastic properties of methylcellulose/agarose hydroge – **B. Niemczyk**, P. Sajkiewicz

19. Magneto-Plasmonic nanoparticles for photothermal therapy – **C. Multari**, M. Miola, F. Laviano, R. Gerbaldo, G. Pezzotti, D. Debellis, E. Vernè

20. Evaluation of mechanical properties and biocompatibility of Gum Metal for implant applications – **K. Golasiński**, E. Pieczyska, R. Detsch, A.R. Boccaccini, N. Takesue

Poster session / Buffet dinner (19:15-21:00)

Workshop programme

28th September 2017

Registration and coffee (8:30-9:00)

Keynote lectures III (9:00-11:00) – *Chair*: Aldo R. Boccaccini (FAU)

- Cartillage tissue engineering with biomaterials developed for cartilage repair – **Gundula Schulze-Tanzil** (Institute for Anatomy, Klinikum Nürnberg Medical School)
- Recent multiscale mechanics contributions to bone tissue engineering – **Christian Hellmich** (TU Vienna)
- Surgery and tissue engineering. What is the role of vascularization? – **Raymund Horch** (University Hospital Erlangen)
- Mimicking Bone Technology: Transforming implant surfaces from an artificial barrier into a smart implant body interface – **Dietmar Schaffarczyk** (stimOS GmbH)

Coffee break (11:00-11:30)

Oral session II (11:30-13:00) – *Chair*: Heike Walles (University of Würzburg)

Ex vivo osteochondral explant model: Creation of standardized defects to investigate cartilage treatment strategies – **A. Schwab**, A. Buß, S. Naczenski, H. Walles, F. Ehlicke

3D bioprinting of chondrocyte-laden CELLINK hydrogels for patient-specific auricular cartilage reconstruction – **S. Schwarz**, H. Martínez Ávila, N. Rotter, P. Gatenholm

Surface patterning of a novel functionalized Poly-L-lactide polymer to improve its biocompatibility: Applications to Bioresorbable Vascular Stents (BVS) – S. Pacharra, **R. Ortiz**, S. McMahon, W. Wang, J. Salber, I. Quintana

Pressureless spark plasma–sintered Bioglass® 45S5 with enhanced mechanical properties and stress–induced new phase formation – **L. Bertolla**, I. Dlouhý, P. Tatarko, A. Viani, A. Mahajan, Z. Chlup, M.J. Reece, A.R. Boccaccini

Production of ATMPs under the regulatory requirements – **Ch. Rücker**, M. Haddad-Weber, P. Bittdorf, H. Walles, O. Pullig

3D investigation and visualization of biomaterials at micro- and nanoscale – **J. Karbowniczek**, S. Metwally, P. Szewczyk, A. Gruszczyński, A. Kruk, A. Czyrska-Filemonowicz, U. Stachewicz

KMM-VIN Forum & Networking Session (13:00-13:30)
Aldo R. Boccaccini and Michał Basista

Buffet lunch/End of Workshop (13:30-14:30)

Abstracts: Keynote lectures

Keynote lectures

Functionalization of surfaces and applications in healthcare surroundings

Frank Heidenau

BioCer Entwicklungs GmbH
Bayreuth, Germany

Sol–gel chemistry offers a flexible approach to obtaining different materials as well as a wide range of nano-/micro-structures that can be used for coating technology. The low processing temperatures combined with an intrinsic bio-compatibility of the commonly used components makes it an ideal method for biomedical coating applications.

During this lecture, an overview of some applications for sol-gel derived coatings in medical applications is presented. This covers surface modifications of orthopedic and dental implants as well as concepts about improvements in the clinical environment of patients like novel cleaning systems or antibacterial surfaces.

The proposed surface modifications cover biocompatible, antibacterial and osteointegrative properties of coatings as well as combinations of the single functionalizations.

Opportunities and challenges of 3D printing in healthcare applications

Tim Van Cleynenbreugel

3D Systems Leuven
Grauwmeer 14, 3001 Leuven, Belgium

3D printing, also called Additive Manufacturing, was invented in 1983 by Chuck Hull, the founder of 3D Systems. Initially it was only used for prototyping. However, 3D printing is now at an infliction point where it is shifting from prototyping to production. Also the medical device industry is rapidly adopting 3D printing technology, both in polymers and in metals, where it enables the manufacturing of complex organic shapes and geometries. These in turn allow for new or improved treatments using 3D printed models, instruments or implants with advanced designs and characteristics. Typical examples include patient specific anatomical models and surgical guides, and metallic implants with complex shapes and porous structures for optimal bone ingrowth.

In this presentation, the state-of-the-art of 3D printing in healthcare is presented. A complete overview of different technologies is given. Technologies for polymer printing include Stereolithography (SLA), Selective Laser Sintering (SLS), ColorJet Printing (CJP), MultiJet Printing (MJP), and Direct Light Projection (DLP).

The remainder of the presentation will focus on Direct Metal Printing (DMP). In DMP a laser beam is used to melt fine metal powder in a layer-by-layer fashion, resulting in fully functional metal parts. Typical metals for DMP of medical implants include titanium and its alloys (Ti Grade 1, 5 and 23), and Cobalt-Chrome (CoCr ASTM F75). Surgical instruments are often printed in Stainless Steel (316L or 17-4 PH). During the presentation the challenges and opportunities for 3D metal printing in healthcare will be discussed, and illustrated with real life examples.

Requirements for ceramic implants for hip joint prostheses

Thomas Oberbach

Mathys Orthopaedie GmbH
An den Trillers Bueschen 2, 07646 Moersdorf, Germany

thomas.oberbach@mathysmedical.com

The pure ceramic material Al_2O_3 as ceramic material for hip joints was introduced into the market in 1969/1970. Beside the alumina ceramic in 1985 ZrO_2 (Y-TZP) was introduced. But the big disadvantage of pure Y-TZP is its lower stability against low temperature degradation (LTD). This is combined with an increase of the surface roughness and a decrease of the strength.

A way of combining the positive properties of Al_2O_3 (wear resistance, hydrothermal stability and hardness) with those of ZrO_2 (strength and fracture toughness) are the dispersion ceramics ATZ and ZTA. Due to the higher strength of the dispersion ceramics by their use for standard applications in hip endoprosthetics the ceramic associated problems like in vivo fracture rate can be significantly decreased.

The wear behaviour of ZTA and ATZ is much better compared to the pure ceramics Al_2O_3 and ZrO_2 (Y-TZP). With an ATZ combination for hip heads and cups the wear in a hip simulator using microseparation can be reduced 12 times compared to Al_2O_3. So the risk for ceramic associated squeaking is reduced.

Actual tendencies on the market show that ZTA is suitable for standard implants and will continuously replace the alumina components. ATZ is the best solution for high-performance demands with complex geometries and high reliability. So the use of the material for knee prosthesis are ceramic resurfacing components can be possible.

The presentation will compare the properties of the dispersion ceramics ZTA and ATZ and show activities for new ceramic implants for joint endoprostheses.

Soluble phosphate glasses and composites as key biomaterials

David M. Healy

IDP Services Limited, UK
Midton House, By Alloway, Ayr, Scotland KA7 4EG

david@idp.services

The presentation provides a broad overview of soluble phosphate glasses and their inclusion in hydrogel and biodegradable polymer composites. Included is a brief description of phosphate glasses and how they differ from bioglass compositions.
Their versatility as key biomaterials is exemplified by presenting how these materials can be manipulated and produced for uses in nine of the eleven topics indicated in the Workshop flyer "Biomaterials: Key Technologies for Better Healthcare". This includes development of materials for use as:

- Biomaterials for bone and dental implants
- Biomaterials for cardiovascular applications
- Bone filling materials
- Scaffolds for hard and soft tissue engineering
- Bioactive and cell instructive materials
- Drug delivery carriers
- Biomedical Coatings
- 3D biomaterial models for cancer research
- Clinical performance of biomaterials

When cell meets textiles – towards biohybride implants

Stefan Jockenhövel

NRW-Schwerpunktprofessur Biohybrid & Medical Textiles (BioTex) at:
Institute for Applied Medical Engineering, Helmholtz Institute Aachen, Germany
Institut für Textiltechnik, RWTH Aachen University, Germany
Aachen-Maastricht Institute for Biobased Materials, Maastricht University, The Netherlands

jockenhoevel@hia.rwth-aachen.de

Regenerative medicine is a research area has promised the paradigm shift from fully technical, avital implants towards vital implants based on the patients' own cells. Although many preclinical studies have been successful demonstrated the feasibility of such implants, just a limited number have reached the level of clinical practice.

There are a number of reasons, which are making the translation of pure biological implants into the clinic so problematic. First of all the cell behaves like a complex adaptive system. The control of such a fully complex adaptive systems towards a robust production process is extremely difficult. Therefore the optimal combination (golden cut) of technical and biological components is very promising (= biohybrid implants). Combining the best of both a (i) robust (reproducible) production process and a high (bio)mechanical performance by the technical component and the (ii) physiological hemo- and biocompatibility of the biological component will help to overcome the current limitations in implantology.

Why are textiles herefore so interesting? The biomechanical properties of the human body are mainly defined by fibre structures (collagen, elastin, fibrin, fibrose cartilage ect.). So textile Engineering offers a multi-scale toolbox for the development of biomimetic structures on (1) the molecular level of polymer science and biochemical functionalisation, (2) the nano/micro-scale level of fibre production (e-spinning, melt-, wet-, dry-spinning) including the incorporation of drugs and markers and on a (3) meso/macro-scale level for the production of 2D and 3D structures by weaving, knitting, braiding etc.

The lecture will give an overview about the different textile technologies and their impact for tissue engineering in general and in thoracic & cardiovascular tissue engineering specifically.

LIPUS – Low Intensity Pulsed Ultrasound for bone healing purposes: Only a gimmick or a mechanism-based bone regenerative approach

Jochen Salber

University Hospital Knappschaftskrankenhaus Bochum
Bochum, Germany

30 years ago the first level one clinical trial about the application of LIPUS on fractured bone was carried out. This study could demonstrate an acceleration of fracture repair. But the underlying physiological processes and mechanisms remain unclear.

An aging population leads in traumatology as well to new problems. Fracture healing with respect to comorbidities like osteoporosis and /or diabetes 2 are challenging orthopedic surgeons all over the world. Understanding the stimulating character of LIPUS would help to optimize bone fracture healing or bone regeneration respectively. Especially the combination of LIPUS with optimized regenerative biomaterials containing the necessary structure-function-relationship could reduce the time of postoperative immobility of the older patient. I would like to highlight the current knowledge about the mechanotransductive processes occurring during LIPUS application to cells of different origin.

Cartilage tissue engineering with biomaterials developed for cartilage repair

Gundula Schulze-Tanzil

Department of Anatomy, Paracelsus Medical University, Salzburg and Nuremberg, Germany

Cartilage tissue has only a very limited intrinsic repair capacity. Therefore, the treatment of injured cartilage remains highly challenging. Various cartilage tissue engineering (TE) strategies have been established to manufacture artificial neocartilage for transplantation to address this unmet medical need. Biomaterial-free approaches and several types of scaffolds have been developed for cartilage TE.

However, satisfying repair of damaged hyaline cartilage could not be attained so far. Biomimetic scaffolds combined with an optimized functionalization are still required to promote chondrocyte or progenitor cell growth and chondrogenic differentiation to facilitate cartilage repair. Hence, the question arises, which biomaterial manufacturing, functionalization and cell seeding strategies could indeed promote chondrogenesis.

In cooperation with several research groups of material scientists various porous, fibrous and hydrogel-based scaffold types and functionalization strategies were tested by our team for their effects on chondrogenesis in view of applicability for cartilage repair. An important topic was to identify biomaterial- and cell-based influence factors important for cartilage TE to optimize future approaches for cartilage repair.

PLLA scaffolds of varying pore sizes prepared by thermally induced phase separation (TIPS) and functionalized or not with bioactive glass (BG) were seeded with human chondrocytes of different origin or with human mesenchymal stromal cells (MSCs). In addition, a modified hydrogel and a newly developed pure BG scaffold were tested.

Highly porous PLLA scaffolds and modified hydrogels allowed high cell survival and chondrogenic marker expression of MSCs, articular and nasoseptal chondrocytes presenting an interesting basis for future modifications. Scaffolds with smaller pore sizes (100 µm) led to superior results compared with those with larger pores (200 µm). Chondrocytes exposed to BG maintained their differentiated phenotype. First insights revealed that BG, including a novel BG type, directly adapted to chondrocytes, could present a promising future functionalization approach to create multiphasic composite scaffolds for osteochondral repair.

Keywords:

Cartilage Tissue Engineering; Articular and nasoseptal chondrocyte, mesenchymal stromal cells, PLLA, bioglass.

Recent multiscale mechanics contributions to bone tissue engineering

Christian Hellmich

Institute for Mechanics of Materials and Structures
TU Vienna, Austria

Bone tissue engineering materials must blend in the targeted physiological environment, in terms of both the materials' biocompatibility and mechanical properties. As for the latter, a well-adjusted stiffness ensures that the biomaterial's deformation behavior fits well to the deformation behavior of the surrounding biological tissue, whereas an appropriate strength provides sufficient load-carrying capacity of the biomaterial. Here, a mathematical modeling approach for estimating the macroscopic load that initiates failure of a hierarchically organized, granular, hydroxyapatite-based biomaterial is presented. For this purpose, a micromechanics model is developed for downscaling macroscopically prescribed stress (or strain) states to the level of the needle-shaped hydroxyapatite crystals. Presuming that the biomaterial fails due to the quasi-brittle failure of the most unfavourably stressed hydroxyapatite needle, the downscaled stress tensors are fed into a suitable, Mohr-Coulomb-type failure criterion, based on which the macroscopic failure load is deduced. The change of the biomaterial's composition in response to placing it in physiological solution, caused by growth of new bone tissue on the granules's surfaces, on the one hand, and by resorption of the hydroxyapatite crystals, on the other hand, is taken into account by means of suitable evolution laws. Numerical studies show how the macroscopic load carrying capacity of the biomaterial is influenced by its design parameters. The presented modeling approach could prove beneficial for the design process of the studied biomaterials (as well as similarly composed biomaterials), particularly in terms of optimizing its mechanical performance.

References

Scheiner S, Komlev VS, Hellmich C. Int J Fract (2016) 202:217–235

Scheiner S, Komlev VS, Gurin AN and Hellmich C (2016) Front. Physiol. 7:383. doi: 10.3389/fphys.2016.00383

Dejaco A, Komlev VS, Jaroszewicz J, Swieszkowski W, Hellmich C (2016) Bioinspired, Biomimetic and Nanobiomaterials 5(1): 24–36, http://dx.doi.org/10.1680/jbibn.15.00021

Surgery and tissue engineering. What is the role of vascularization?

Raymund E. Horch

University Hospital Erlangen

Department of Plastic and Hand Surgery
Krankenhausstrasse 12, 91054 Erlangen, Germany

Tissue Engineering (TE) and Regenerative Medicine (RM) are promising tools to overcome current unmet clinical needs with regard to tissue and organ replacement. Despite considerable advances in material and biomaterial design and fabrication techniques, however, it has not entered the clinical arena yet. With few exceptions and anecdotal reports TE and RM are not available for daily routine so far. One problem has been the lack of initial vascularizarion of TE substitutes in clinically relevant sizes. Nevertheless a functional blood vessel network is a prerequisite for the survival and growth of almost all tissues and organs in the human body. Moreover, in pathological situations such as cancer, vascularization plays a leading role in disease progression. Consequently, there is a strong need for a standardized and well-characterized in vivo model in order to elucidate the mechanisms of neovascularization and develop different vascularization approaches for tissue engineering and regenerative medicine.

We describe a microsurgical approach for a small animal model for induction of a vascular axis consisting of a vein and artery that are anastomosed to an arteriovenous (AV) loop. The AV loop is transferred to an enclosed implantation chamber to create an isolated microenvironment in vivo, which is connected to the living organism only by means of the vascular axis. Using 3D imaging (MRI, micro-CT) and immunohistology, the growing vasculature can be visualized over time. By implanting different cells, growth factors and matrices, their function in blood vessel network formation can be analyzed without any disturbing influences from the surroundings in a well controllable environment. In addition to angiogenesis and antiangiogenesis studies, the AV loop model is also perfectly suited for engineering vascularized tissues. After a certain prevascularization time, the generated tissues can be transplanted into the defect site and microsurgically connected to the local vessels, thereby ensuring immediate blood supply and integration of the engineered tissue. By varying the matrices, cells, growth factors and chamber architecture, it is possible to generate various tissues, which can then be tailored to the individual patient's needs. This approach highlights the relationship between surgical techniques and TE research.

Mimicking Bone Technology: Transforming implant surfaces from an artificial barrier into a smart implant body interface

Dietmar Schaffarczyk

stimOS GmbH
Byk-Gulden-Straße 2 // F21, D-78467 Konstanz, Germany

Abstract. Surgeons and patients, as well as medical device manufacturers are frequently confronted with post-operative implant failures due to implant loosening or inflammatory reactions. These complications are often the reason for pain after surgery, and lead to revision surgeries resulting in increased healthcare costs.

These failures are not limited to certain indications or surgical techniques: They occur in all applications where implants must be placed in the patient's body.

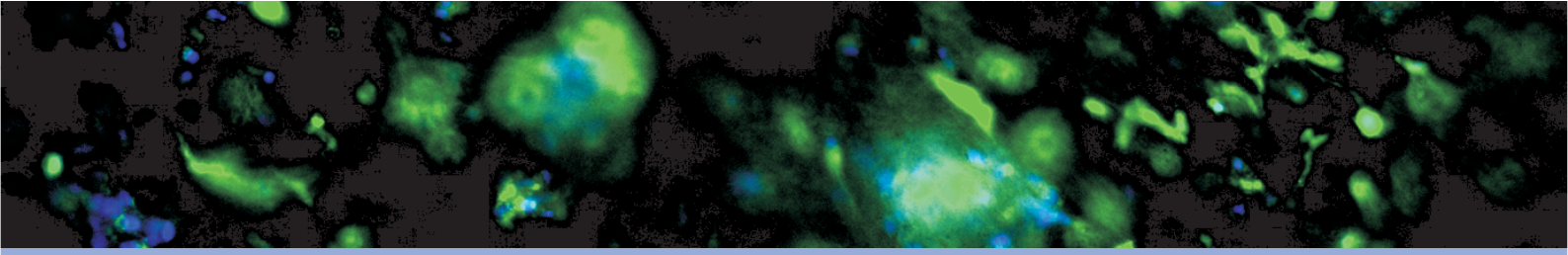
Reasons for failed surgeries can be: [1] Implant materials available have not the best biologic performance due to their material characteristics. [2] The surrounding bone – where the surgeon has to place the implant – is not stable or dense enough due to the patient's age and/or osteoporotic skeletally bones. As surgeons are forced to deal with such poor conditions, medical device manufacturers shall enhance the performance of implant surfaces, to achieve more natural material characteristics.

With its Mimicking Bone Technology (MBT), stimOS GmbH aims to modify the implant surface from an artificial barrier in the patient's body to a smart implant body interface. MBT operates as a „stealth technology" for implants, transforming a neutral implant surface into a bone- identic implant body interface.

With MBT, stimOS offers the surgeon and the patient a completely new solution. The company opens new, innovative paths by modifying the surface structure of implant materials biochemically by means of a covalently bound activation layer, rather than relying on coating technologies. stimOS' MBT gives even inert materials biological properties close to nature and similar to those found naturally in the human body.

About stimOS

stimOS GmbH was founded in May 2015 by Prof. Dr. Helmut Cölfen, Prof. Dr. Günter Schatz, Prof. Dr. Johannes Boneberg and Dr. Dietmar Schaffarczyk. stimOS develops innovative technologies and procedures that refine, functionalize and activate implant materials. As a supplier and service provider, stimOS makes these technologies available to implant manufacturers. Furthermore, the company offers services in the field of product development and certification, and develops with the product line spineFuseMBT implants for spinal fusion surgery.



Abstracts: Oral sessions

Surface modifications at the nanoscale for a better integration of implants to the hard and soft tissues and a reduced bacterial contamination

S. Spriano [1], F. Baino [1], C. Balagna [1], M. Cazzola [1], A. Cochis [3], M. Ferraris [1], S. Ferraris [1], M. Miola [1], C. Ramskogler [4], L. Rimondini [3], A. Varesano [2], E. Vernè [1], C. Vineis [2], F. Warchomicka[4]

[1] *Politecnico di Torino, Italy*

[2] *ISMAC-CNR, Biella, Italy*

[3] *Università del Piemonte Orientale, Novara, Italy*

[4] *Graz University of Technology, Graz, Austria*

Tissue integration of biomaterials can be increased by several modification at the nanoscale: we explored nanoparticles precipitation, thin films, nanostructuring of the surface and nanofiber deposition.

Through nanoparticle precipitation and thin films antibacterial agents can be added onto surfaces: silver nanoparticles and nanoclusters were investigated. Nanoparticles precipitation was obtained by chemical processes while nanoclusters were embedded in a silica matrix during sputtering deposition on different substrates. Antibacterial activity and ion release were monitored.

Nano-structuring of the surface can be useful in order to get mineralization ability (apatite precipitation) of the surface, as well as better cell differentiation (osteoblasts) or orientation (fibroblasts). In this way, the integration of the implant can be increased because of faster growth of bone or guided growth of oriented soft tissue. Surface nanostructuring can be coupled with modified surface chemistry, such as by high hydroxylation. Titanium surfaces with a nanogrooved (oriented) or sponge-like (random) structure have been analysed. In-vitro apatite precipitation and cell response were tested.

Nanofiber deposition can enhance and guide soft tissues adhesion (e.g. gum). Moreover nanofibers can be enriched with metallic ions for antibacterial properties. Biological (bacterial and cell cultures) and chemical tests were performed.

All the obtained results are discussed in view of understanding potential applications of nanoscale modifications of biosurfaces in dental and orthopaedic implants.

Site-directed immobilization of BMP-2 onto collagen beads for bone regeneration

Claudia Siverino[1,2], Joachim Nickel[1,2], Heike Walles[1,2]

[1] *Translational Center Würzburg 'Regenerative therapies in oncology and musculoskeletal disease' Würzburg branch of the Fraunhofer Institut Interfacial Engineering and Biotechnology (IGB)*

[2] *Department of Tissue Engineering and Regenerative Medicine, University Hospital Würzburg Germany*

The use of biomaterials and growth factors has been the focus of bone tissue engineering for the treatment of non-healing long bone defects. Current approaches are based on the incorporation of an osteogenic growth factor, the bone morphogenetic protein 2 (BMP2) into the scaffold. However, the common used methods, such as surface absorption and encapsulation, require high doses of protein which are associated to several severe side effects. In our study we aim to overcome these problems by covalent immobilization of BMP2 in a site directed manner. For this purpose, we designed two BMP2 variants harboring one unique non-natural amino acid substitution in each chain of the mature polypeptide allowing a site-specific coupling by "click chemistry". The BMP2 variants were expressed in E.coli and purified from inclusion bodies. The final products showed the same bioactivity as the wildtype BMP2. Using either copper catalyzed or copper free click chemistry reactions, the proteins could be successfully coupled to collagen microspheres. The efficiency of the coupling reactions was validated with different analytical methods. The final prove of the efficacy of this new approach is now being evaluated in vivo in an animal experiment. Using a paste composition of the collagen microspheres, we injected different doses of BMP2 variants subcutaneously using absorbed BMP2 wildtype as control. Micro-computed tomography (µCT) analysis revealed bone formation at the injection site at week 4. Evaluation of bone volume and density will be performed at different time points until the end of the animal experiment when implants are histologically analyzed. The covalently coupled BMP2 approach might serve as an innovative biomaterial with superior bone healing properties for clinical applications.

Electrospun multilayered scaffolds for interface tissue engineering applications

Liliana Liverani* and Aldo R. Boccaccini

Institute of Biomaterials, Department of Materials Science and Engineering

University of Erlangen-Nuremberg, Cauerstr. 6, 91058, Erlangen, Germany

** liliana.liverani@fau.de*

The fabrication of scaffolds able to replicate the native tissue anisotropy has been the focus of recent research efforts for the regeneration of interface tissues, like osteochondral segment. The electrospinning technique is widely investigated for the fabrication of fibrous scaffolds able to replicate the morphology of the native extra cellular matrix. The use of benign solvents for the electrospinning has numerous advantages, because it avoids the denaturation of proteins and biomolecules, eliminates the presence of toxic solvent residuals and has positive impact on lab workers safety and waste management.

In the present work, the electrospinning with benign solvents was used for the fabrication of scaffolds with gradients in composition and porosity. In fact these multilayered scaffolds were obtained with consequent electrospinning of different polymeric solutions and blends. The selected polymers were poly(epsilon-caprolactone) (PCL) and chitosan. PCL was selected for its electrospinnability, biocompatibility and biodegradability and chitosan was selected for its biocompatibility, antibacterial properties and effects on chondrogenesis. Composite electrospun fibers were obtained by incorporation of bioactive glass (BG) particles in the polymeric solutions before the electrospinning process. Gradients in composition were obtained by electrospinning on polymeric layer and composite layer.

Positive results were obtained from the morphological, chemical and mechanical characterization performed on the single layer and multilayered samples. Bioactivity assessment was also performed on composite monolayer and on the multilayered sample with composite layer, confirming the preservation of BG bioactivity even if incorporated in the polymer mat.

Preliminary in vitro tests with bone marrow stromal cells were performed on single layer scaffolds and multilayered scaffolds to investigate cell viability and morphology.

Influence of polymer film concentration on cytocompatibility and corrosion suppression of ZM21 magnesium alloy

Agnieszka Witecka[1,2,3], Akiko Yamamoto[1], Wojciech Święszkowski[2], Michał Basista[3]

[1]*National Institute for Materials Science, Tsukuba, Japan*

[2]*Warsaw University of Technology, Warsaw, Poland*

[3]*Institute of Fundamental Technological Research, Polish Academy of Sciences, Warsaw, Poland*

Mg alloys are promising materials for biodegradable medical devices. However, control of Mg alloys corrosion is crucial for their success as implant devices. Application of biodegradable polymer film can improve initial cytocompatibility and corrosion resistance of Mg substrate.

In this work, effect of different biodegradable polymers and their concentrations on films properties were investigated in terms of cytocompatibility and corrosion protection. Poly-L-lactide(PLLA), poly(3-hydroxybutyrate)(PHB) and poly(3-hydroxybutyrate-co-3hydroxyvalerate)(PHBV) were utilized to prepare polymer films with concentration 1%(w/v) and 2%(w/v) on cast Mg-2.0Zn-0.98Mn (ZM21) magnesium alloy by spin-coating method. The main difference between polymers are molecular weight, crystallinity, hydrophobic moieties and degradation rate[1].

WST-1 results confirm SaOS-2 cell growth on PLLA, PHB and PHBV coated samples, however different polymer concentration is reflected in cells viability (Fig.1a). Films from 2%(w/v) concentration indicate higher cell growth than those with 1%(w/v). After 7d of incubation, PHBV 2% tended to be the most beneficial in cell growth improvement, and following are PHB 2%(w/v) and PLLA 2%(w/v). PLLA 1%(w/v) has the lowest contribution in cell growth. Mg^{2+} ion release was observed for all samples and increased along incubation period (Fig.1b). Slightly difference between PLLA and PHAs (polyhydroxyalkanoates: PHB and PHBV) was noticed at 7d, indicating PHAs as more beneficial in suppression of Mg^{2+} release.

Obtained results can be related to slightly thicker 2%(w/v) films and polymer physicochemical properties itself.

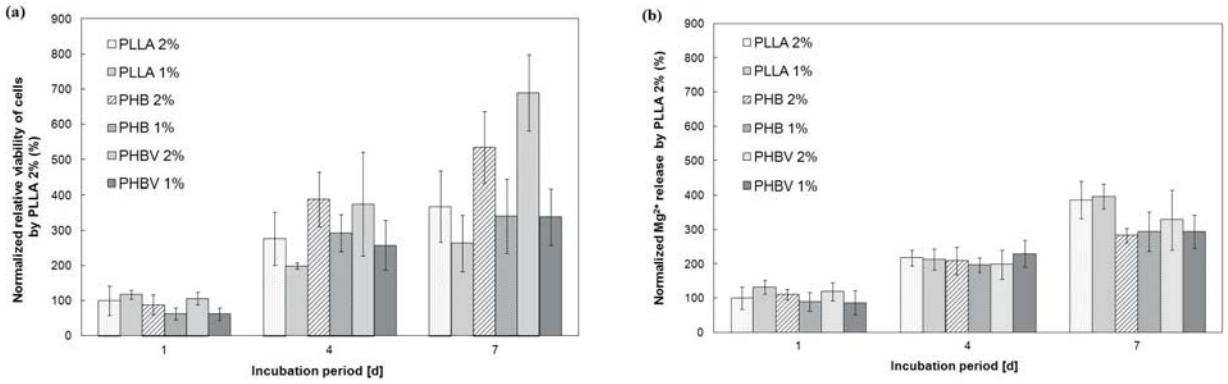


Fig.1 (a) Normalized data by PLLA 2%(1d) for polymer coated samples, (b) relative viability of cells Mg^{2+} ions release.

[1] A. Witecka, A. Yamamoto, J. Idaszek, A. Chlanda, W. Święszkowski (2016), Colloids Surf. B 144: 284-292

Hybrid materials for biomedical applications

J. Carlos Almeida, Isabel M. M. Salvado, M. Helena V. Fernandes

Department of Materials and Ceramic Engineering, CICECO, Universidade de Aveiro
3810-193 Aveiro, Portugal

The hybrid system PDMS-SiO_2 is apparently simple. In fact the $Si(CH_3)_2$-O-$Si(CH_3)_2$ arrangement in the PDMS molecule closely resembles the silica backbone and there is a great facility in obtaining PDMS hyrdroxyl terminated forms. On the other hand, PDMS (usually known as silicone) has been used for a long time in biomaterials with good results and extensive record. Almost naturally the two compounds met in the first studies on hybrid materials. The possibilities open by joining other oxides to this binary system in order to obtain materials for different applications, from biomaterials to photonics, creates the need for a better understanding of the processing-structure-properties relationships. The present work deals with these relationships, some of them initially approached by other authors, with the focus on the biomedical applications, but also expecting to create some windows for other domains.

Titanium and zirconium are known for their ability to improve bioactivity, but they also strongly affect the structural and microstructural features of the SiO_2-TiO_2 and SiO_2-ZrO_2 binary systems, and also of the PDMS-TiO_2 and PDMS-ZrO_2 systems. Detailed studies with different sol-gel conditions allowed the understanding of the roles of titanium and zirconium as additives in the PDMS-SiO_2 system. It was concluded that titanium and zirconium influence the kinetics of the sol-gel process due to their different alkoxide reactivity leading to hybrid xerogels with dissimilar characteristics and morphologies. The possibility of modulating the structure and microstructure of these hybrids was seen to open interesting alternatives for the production of cytocompatible scaffolds adapted to different tissue engineering situations.

Single cell analysis for studying tumor heterogeneity

Lucas Hoene[1], Andreas Schwab[1], Ben Fabry[1], Manuel Théry[2], Aldo R. Boccaccini[1], Aldo Leal-Egana[1]*

[1] *Institute of Biomaterials, University of Erlangen-Nuremberg, Germany*

[2] *CEA Research Institute, Paris, France*

aldo.leal.egana@fau.de

Biomaterials mimicking the neoplastic milieu have become a straightforward alternative for assaying cancer progression, tumor heterogeneity and for testing cell pathogenicity in vitro.

Recently, we analyzed the migration of single cells (healthy and metastatic breast cells) on soft polyacrylamide hydrogels patterned with thin migration guidelines (4µm width), to characterize their metastatic capability.

These scaffolds resembled the fibrillar topology of ECM proteins found within the tumoral niche, mimicking the mechanical and topographical cues found in tumoral environments. Using these biomimetic platforms, we performed time-lapse analysis of cell morphology, migration speed and traction forces exerted by single-cells.

According to our results, wild-type cells (MCF10A; non-metastatic) are characterized by the existence of two sub-populations within this phenotype, differentiated in terms of length (round or large cells), speed (fast or slow cells), and exertion of traction forces (strong or weak cells).

Interestingly, this behavior is shifted in the case of highly metastatic cells (i.e. MCF10A-ErbB2), or those stimulated with growth factors to acquire a pathogenic behavior (i.e. MCF10A cultured in presence of TGF-β), being defined by the existence of a predominant population, characterized by its small and round morphology, fast migration speed, and generation of low mechanical forces.

It is important to note that these results cannot be assessed by averaging experimental data obtained from an entire population, or considering all cells as a whole. Thus, in this work we remark that the metastatic potential of healthy/cancerous phenotypes may not be determined in vitro by averaging their behavior, or analyzing them as a homogeneous group of cells, rather than extracting tendencies from assays performed at single-cell scale, pondering the potential existence of cell sub-types within a multicellular experiment1.

References

1. Leal-Egana et al., (2017). The size-speed-force relationship governs migratory cell response to tumorigenic factors. Molecular Biology of the Cell. 28:1612-1621

Ex vivo osteochondral explant model: Creation of standardized defects to investigate cartilage treatment strategies

A. Schwab[1], A. Buß[1], S. Naczenski[1], H. Walles[1,2], F. Ehlicke[1]

[1] *University Hospital Würzburg, Tissue Engineering & Regenerative Medicine, Würzburg, Germany*

[2] *Translational Center Würzburg ´Regenerative Therapies in Oncology and Musculoskeletal Disease`, Würzburg, Germany*

Since cartilage defect treatment is dependent on lesion severity, pre-clinical cartilage models need to address clinical relevant defect geometries ranging from fissures and cartilage only lesions to osteochondral defects. Aim of this study was to modify the ex vivo osteochondral explant (OCE) model[1] by creating standardized cartilage defects, different in depth. Matrix assisted cell free as well as cell loaded treatment approaches were compared to investigate the influence of defect depth and oxygen tension on cartilage regeneration in ex vivo model.

OCE were isolated from porcine femoral condyle (diameter: 8mm, height 5mm). Full thickness cartilage defects (diameter: 4mm) were created with biopsy punch. Chondral defects, 1mm in depth, were induced with the automated device Artificial Tissue Cutter (ARTcut®). Lesions were either left untreated or filled with cell free or chondrocyte loaded collagen I hydrogel and cultured for 4 weeks under static conditions. Separated media compartments allowed to supply explant with tissue specific nutrients. Live-dead staining was performed to investigate cell viability and cartilage regeneration was evaluated by (immuno-) histological stainings and quantification of proteoglycans (GAG) in hydrogels.

Cells hosted in OCE and chondrocytes seeded in hydrogel were viable after 4 weeks culture. Cell invasion into cell free hydrogel was observed originating from surrounding cartilage of OCE. No intrinsic self-healing of defects was achieved in cell free hyrogels. Chondrocytes in cell loaded approach synthesized cartilaginous matrix, positive for collagen II, aggrecan and SOX9 with absence of collagen I and X.

Separated media compartments with tissue specific nutrient supply is critical in preserving matrix composition and vitality during ex vivo culture. OCE model represents a pre-clinical tool to evaluate novel scaffold materials on their regenerative capability. ARTcut® is an innovative wounding device to mimic different lesion severities and thus study degenerative mechanisms and regenerative treatment strategies ex vivo.

References

1. A. Schwab, A. Meeuwsen, F. Ehlicke, J. Hansmann, L. Mulder, A. Smits, H. Walles and L. Kock; 'Ex vivo culture platform for assessment of cartilage repair treatment strategies', ALTEX, 2017, vol. 34, no. 2, p.267-277, doi.org/10.14573/altex.1607111. Epub 2016 Oct 21.

Acknowledgements

The work leading to these results has received funding from the European Union Seventh Framework Programme (FP7/2007-2013) under grant agreement n° 309962. ARTcut® was developed in cooperation with Fraunhofer ISC (Würzburg).

3D bioprinting of chondrocyte-laden CELLINK hydrogels for patient-specific auricular cartilage reconstruction

Silke Schwarz[1,2], Héctor Martínez Ávila[3,4], Nicole Rotter[2,5] and Paul Gatenholm[2]

[1] *Department of Anatomy, Paracelsus Medical University, Salzburg and Nuremberg, Germany*

[2] *Department of Otorhinolaryngology, Ulm University Medical Center, Ulm, Germany*

[3] *Biopolymer Technology, Department of Chemistry and Chemical Engineering, Chalmers University of Technology, Gothenburg, Sweden*

[4] *CELLINK AB, Gothenburg, Sweden*

[5] *Department of Otorhinolaryngology, Kepler University Clinic, Linz, Austria*

Cartilage tissue engineering (TE) for auricular reconstruction aims to provide an effective and patient-individual treatment for acquired or congenital auricular defects. In recent years 3D bioprinting has gained growing attention in the field of TE since it offers the possibility to produce patient-specific implant matrices. Due to the possibility to control the distribution of cells, biomaterials and bioactive molecules, 3D printing has become an indispensable tool for a wide variety of TE strategies. To date, considerable progress has been made in bioprinting of complex 3D tissue analogues, but the development of bioinks combining excellent printability, biocompatibility and bioactive properties must be promoted for clinical application.

For patient-individual regeneration of auricular cartilage, we evaluated the biological functionality of CELLINK, a bioink consisting of nano-fibrillated cellulose supplemented with 0.5 % alginate. Within this study, CELLINK was laden with human nasal chondrocytes (hNC), auricular constructs were produced by 3D bioprinting and cultured for up to 28 days. The chondrogenic phenotype of hNCs and neo-synthesis of cartilage specific components was examined on protein- as well as on gene-expression level.

3D printing combined with the biologically active bioink CELLINK facilitated the production of cell-laden implants with a high cell density, a homogenous cell distribution and an open inner structure. The high porosity of the constructs provided an optimal nutrition supply of the embedded hNCs, confirmed by the detected high cell vitality and proliferation during 3D culture. All 3D constructs exhibited excellent shape and size stability. Gene expression, immunohistochemical and biochemical analyses revealed that CELLINK supports redifferentiation and chondrogenesis of hNCs.

In this study we demonstrated that the non-cytotoxic CELLINK provides a biologically relevant micro-environment promoting hNC redifferentiation while offering excellent printability and bioactive properties at the same time. These characteristics make CELLINK a promising tool for auricular cartilage TE and many other patient-specific biomedical applications.

Acknowledgments

Vinnova and Eureka are acknowledged for funding the project EUROSTARS E!8355 CELLINK.

Keywords: 3D Bioprinting; Tissue Engineering; Auricular Cartilage; Neo-cartilage; Nano-fibrillated Cellulose; Human nasal Chondrocytes, patient-specific medicine.

Surface patterning of a novel functionalized poly-L-lactide polymer to improve its biocompatibility: Applications to Bioresorbable Vascular Stents (BVS)

Sandra Pacharra[1], Rocio Ortiz[2], Sean McMahon[3], Wenxin Wang[3], Jochen Salber[1], Iban Quintana[2]

[1] *Zentrum für klinische Forschung, Ruhr-Universität Bochum and Universitätsklinikum Knappschaftskrankenhaus, Bochum, Germany*

[2] *Physics of Surfaces and Materials, Fundación IK4-TEKNIKER, Iñaki Goenaga 5, 20600, Eibar, Gipuzkoa, Spain*

[3] *Vornia Biomaterials Ltd, Laboratory C, Synergy Centre, Tallaght, Dublin, Ireland and The Charles Institute of Dermatology, School of Medicine and Medical Science, University College Dublin, Ireland*

A novel functionalized Poly (L-Lactic acid) polymer was developed for application as a fabrication material for bio-resorbable vascular scaffold (BVS) manufacturing. Picosecond laser ablation technology is a single-step and contactless method for surface microstructuring of any kind of material via fast material removal, which can be easily adapted for micropatterning of tubular or more complex sample shapes, and scaled up by means of micropatterning of metal molds for manufacturing of plastic components series. This versatile and fast microtechnology was applied on the novel functionalized PLLA to generate patterns with different geometry and density and find a pattern configuration able to promote endothelialization on the surface. The patterns generated included parallel micro-grooves with varying width, depth and inter-groove spacing. These were evaluated by cytocompatibility tests using a L929 cell line, endothelial cell adhesion using human cardiac microvascular endothelial cells (HCMECs) and hemocompatibility tests using human blood. We showed that all surfaces tested (non-patterned and patterned) were cyto-compatible and non-toxic. In addition, some of the patterns significantly enhanced endothelial cell adhesion and alignment with respect to the non-patterned surface. The results obtained from the hemocompatibility tests indicated that laser-created patterns favor platelet adhesion and leukocyte activation on the surface, however, further experiments will be performed to investigate platelet adhesion on the patterns when endothelial cell adhesion occurs simultaneously and blood flow conditions are considered.

Keywords: bio-resorbable scaffolds; ultrashort pulsed laser; surface patterning; endothelialization; cardiovascular stents.

Pressureless spark plasma–sintered Bioglass® 45S5 with enhanced mechanical properties and stress–induced new phase formation

Luca Bertolla[1], Ivo Dlouhý[1], Peter Tatarko[2,3], Alberto Viani[4], Amit Mahajan[2], Zdeněk Chlup[1], Michael J. Reece[1], Aldo R. Boccaccini[5]

[1] *Institute of Physics of Materials ASCR, CEITEC IPM, Žižkova 22, 61662 Brno, Czechia*

[2] *Nanoforce Technology Limited, Queen Mary University of London, London E1 4NS, United Kingdom*

[3] *Institute of Inorganic Chemistry, Slovak Academy of Sciences, Dúbravská cesta 9, 84536, Bratislava 45, Slovakia*

[4] *Centre of Excellence, Telč Batelovská 485-6588 56 Telč, Czechia*

[5] *Institute of Biomaterials, Department of Materials Science and Engineering, University of Erlangen-Nuremberg, 91058 Erlangen, Germany*

Commercial Bioglass® 45S5 powder was sintered using spark plasma sintering (SPS) technique without the assistance of mechanical pressure with heating and cooling rate of 100 °C/min, dwell temperature of 1050 °C and dwell time of 30 min. Such route enabled the production of samples exhibiting superior mechanical properties in comparison with Bioglass® sintered in furnace. In particular, flexural strength and fracture toughness reached values close to those of apatite-wollastonite bioceramics, already widely used in clinical applications. The residual stresses implemented by indentation promoted the formation of a new phase in samples sintered by SPS. Complementary use of Raman and energy dispersive spectroscopy (EDS) indicated the phase as sodium carbide and a formation mechanism was proposed.

Production of ATMPs under the regulatory requirements

Christoph Rücker[1], Meike Haddad-Weber[2], Patrick Bittdorf[2], Heike Walles[1,2], Oliver Pullig[1]

[1] *Fraunhofer Translational Center Würzburg 'Regenerative therapies in oncology and musculoskeletal disease', Branch of the Fraunhofer Institute for Silicate Research ISC*

[2] *Department Tissue Engineering and Regenerative Medicine, University Hospital Würzburg*

New therapeutic strategies in regenerative medicine are based on the development of Advanced Therapy Medicinal Products (ATMPs). ATMPs involve products for gene therapy, somatic cell therapy and tissue engineering with or without biomaterials.

The Fraunhofer Translation Center Würzburg (TLZ-Würzburg) offers an infrastructural environment for the development of tissue engineering approaches including specialized facilities, highly trained staff, and expert knowledge to fulfill regulatory requirements. The sophisticated translation from R&D level to GMP compliance is demonstrated on the manufacturing of complex tissues like xenogenic vascularized scaffolds for the treatment of severe tracheal defects (TraVaSc-TERM™). Furthermore, two autologous products for the treatment of cartilage defects are manufactured under the authorization of local and national authorities (N-TEC and N-CAM). N-TEC and N-CAM find their way to human application within the EU project BIO-CHIP. Here, TLZ-Würzburg embedded production in comprehensive quality assurance measures and successfully received manufacturing authorization according Art. 13 German Medicinal Act, recently. The production follows evaluated process steps including nasal tissue procurement, enzymatic isolation of the chondrocytes followed by proliferation, seeding and maturation of the cells on a collagen type I/III scaffold. The efficacy of N-TEC and N-CAM in the repair of focal cartilage defects is currently evaluated in a randomized phase II clinical trial. Four European trial sites are included (Basel, Freiburg, Milan, and Zagreb). The challenging transport logistic of the nasal biopsy and the final product are performed in a validated procedure to meet legal requirements.

Although regulatory hurdles in ATMP production are high, TLZ-Würzburg successfully demonstrates that in an academic environment the translational step from bench-to-bed side is feasible. Our expert knowledge in GMP, GCP, GLP, and GDP allows us to act as an attractive partner for academia and SMI in the field of advanced therapies.

3D investigation and visualization of biomaterials at micro- and nanoscale

Joanna Karbowniczek*, Sara Metwally, Piotr Szewczyk, Adam Gruszczyński, Adam Kruk, Aleksandra Czyrska-Filemonowicz, Urszula Stachewicz

International Centre of Electron Microscopy for Materials Science and Faculty of Metals Engineering and Industrial Computer Science, AGH University of Science and Technology, Al. A. Mickiewicza 30, 30-059 Krakow, Poland

**jkarbow@agh.eu.pl*

Growing market and increasing demand for novel biomaterials is a driving force in medical engineering. New materials and manufacturing methods are developed to address many medical problems. Biomaterials integration with surrounding tissues is essential for their acceptance and performance, therefore their surface properties and architecture were investigated at nano- and macroscale, influencing cells proliferation.

Here, the focused ion beam-scanning electron microscope (FIB-SEM) tomography was applied to visualize the spatial arrangement of investigated biomaterials. The FIB-SEM tomography consists on removing by FIB layer-by-layer the small portions of investigated sample and imaging of the exposed surface by SEM at each stage. Subsequently, the collection of SEM images is used for 3D reconstruction and visualization by specialised software. FIB-SEM tomography is a technique widely used in materials science for example to characterize the microstructure of metallic alloys in 3D. Currently, it is gaining considerable interest in biomedical engineering, allowing us to study the ceramic coating structure on biomedical Ti6Al7Nb alloy or network of nanofibers and their porosities in 3D. The FIB-SEM tomography is able to provide unique information about the cells attachment to applied coatings and nanofibers as well as cells arrangement and spreading according to surface roughness or fibrous scaffolds designs.

Within our studies we showed that FIB-SEM tomography has a great applicability in biomaterials' research providing spatial resolution information about the internal structure as well as cells attachment to ceramic and polymer surfaces, that are crucial for implants and tissue engineering scaffolds.

Acknowledgement

Work was financially supported from the SONATA 8 project granted by National Science Centre in Poland, No 2014/15/D/ST5/02598.

Abstracts:
Short oral presentations

Exploiting piezoelectric polymeric platforms for neuronal tissue engineering

Maria H.V. Fernandes[1], Nathalie Barroca[1], Ana Marote[2], Abílio Almeida[3], Sandra I. Vieira[2]

Odete A. B. da Cruz e Silva[2] , Paula M. Vilarinho[1]

[1] *Department of Materials and Ceramic Engineering, CICECO, Universidade de Aveiro, 3810-193 Aveiro, Portugal*

[2] *Institute of Biomedicine (iBiMED) and Centre for Cell Biology (CBC), Department of Medical Sciences, Universidade de Aveiro, 3810-193 Aveiro, Portugal*

[3] *Department of Physics, Faculdade de Ciências, Universidade do Porto, 4169-007, Porto, Portugal*

In this study, we exploit the potential of PLLA piezoelectric response for neural tissue regeneration, aiming to develop biodegradable platforms. We have produced and tested 3 different PLLA platforms: solvent cast (SC) films, randomly oriented nanofibers (RNF) and aligned nanofibers (ANF). The platforms were subsequently electrically poled in a lab-built corona poling set-up, to maximize their polarization. Non-poled and poled PLLA platforms were analysed to assess their abilities to influence major cellular processes underlying neuroregeneration, including neuroblastoma cells viability and spreading, the alteration of the cellular proteome towards differentiation, and promotion of the neuronal neuritogenic output. Our results show that poled-induced electrically charged platforms have beneficial effects on neuronal-like proliferation and survival, and potentiate neuritic outgrowth. Poled PLLA aligned nanofibers are promising scaffolds to be envisaged as long shelf life therapeutic devices for neural tissue repair.

Personalised 3D-printed smart eyeglasses for physiological monitoring

Oliver Amft, Rui Zhang, Florian Wahl

Chair of eHealth and mHealth, FAU Erlangen-Nürnberg, Germany

We introduce our research activities on printed wearable accessories and hybrid mechanical and electronic systems and describe the requirements for successful wearable embedded systems, highlighting the pertinent challenges resulting from a diversification in mobile and wearable electronics, personalisation needs, and low-volume medical devices markets. in particular, we present projects around smart eyeglasses that use frame-integrated sensors and electronics to provide robust continuous biomedical monitoring with anatomically fitting sensor/electrode positions.

Through head modelling we identified key landmarks for fitting eyeglasses to a human head. The open source human modelling software MakeHuman offers 146 parameters to describe a human head. We selected 26 parameters which influence eyeglasses frame fit and reduced it to three parameters by removing parameters having redundant effects. Our eyeglasses frame CAD model was adapted to include five parameters to adjust fit. The length and distance between temple ends, lens height and width, and nose width can be adjusted.

The skin contact of smart eyeglasses offers various options to create direct body interfaces, for measuring physiological functions as well as interaction. We are investigating conductive materials for long-term direct skin contact and illustrate applications in heart beat and dietary monitoring.

Stem cell labeling with iron oxide nanoparticles in a 3D environment

Maria Steinke[1,2], Teresa Kilian[1,2], Cordula Grüttner[3], Stephan Hackenberg[4], Florian Fidler[5], Karin Schütze[6], Heike Walles[1,2]

[1] *Fraunhofer Institute for Interfacial Engineering and Biotechnology IGB, Translational Center "Regenerative Therapies for Oncology and Musculoskeletal Diseases"– Würzburg Branch, Röntgenring 11, 97070 Würzburg, Germany*

[2] *University Hospital Würzburg, Chair of Tissue Engineering and Regenerative Medicine, Röntgenring 11, 97070 Würzburg, Germany*

[3] *Micromod GmbH, Friedrich-Barnewitz-Straße 4, 18119 Rostock, Germany*

[4] *University Hospital Würzburg, Department of Oto-Rhino-Laryngology, Plastic, Aesthetic & Reconstructive Head & Neck Surgery, Josef-Schneider-Straße 11, 97080 Würzburg, Germany*

[5] *Fraunhofer Institute for Integrated Circuits IIS, Am Hubland, 97074 Würzburg, Germany*

[6] *CellTool GmbH, Am Neuland 1, 82347 Bernried, Germany*

Tissue-engineered constructs represent a promising approach in the field of regenerative medicine. Using 3D tissue equivalents loaded with human mesenchymal stem cells (hMSCs) damaged tissue could be repaired. To analyze cell behavior and tissue healing non-invasively cells can be labeled with iron oxide nanoparticles and monitored applying magnetic resonance or magnetic particle imaging. Using our most promising iron oxide nanoparticle type (perimag®), we analyzed its biocompatibility in vitro.

Cell labeling and subcellular localization of perimag® were verified by Prussian blue staining and transmission electron microscopy, respectively. Cell viability after labeling was determined using fluorescence-activated cell sorting and genotoxic effects were investigated using the chromosomal aberration test. Cell labeling maintenance was documented up to 6 weeks in two- and three-dimensional culture conditions. The impact of hMSC labeling on proliferation was quantified applying Ki67 immunofluorescence. Magnetic particle spectroscopy (MPS) and Raman spectroscopy served to investigate cell-particle interaction non-invasively.

After labeling almost 100% of hMSCs incorporated perimag®. Whereas in 2D culture the percentage of labeled hMSCs rapidly decreased, in 3D conditions about 20% of the cells were still perimag®-positive 6 weeks later. In 3D conditions cell proliferation was lower in both perimag®-labeled cells and non-labeled controls 7 and 21 days after labeling compared to 2D conditions. The percentage of Ki67-positive hMSCs did not vary between perimag®-labeled and non-labeled cells, neither in 2D nor in 3D conditions. Cell viability after labeling was always above 85% and no genotoxic effects of perimag® were observed. Non-invasive analysis of cell-particle interaction in vitro was realized using MPS. Raman spectra of cell-compatible perimag®-labeled cells significantly differed from spectra of cells treated with a harmful nanoparticle type. Our data show that perimag® allow safe hMSC labeling. The hMSC-loaded, 3D tissue-engineered construct could serve as a graft for regenerative therapies, in which perimag®-labeled hMSCs can migrate to their target.

Wedge-shape compression of MSC-laden Col I gel for meniscus tissue engineering

Antje Kremer*, Jenny Reboredo, Heike Walles

Chair of Tissue Engineering and Regenerative Medicine (TERM), University Hospital of Würzburg

Würzburg, Germany

** Antje.Kremer@uni-wuerzburg.de*

Meniscal injuries result in the deterioration of meniscal tissue and consequently of articular cartilage. The healing potential of the avascular region of the meniscus is limited and no optimal treatment strategy is available. Replacing torn menisci by a tissue-engineered 3D meniscus model seems to be a promising strategy. A 3D meniscus model needs an appropriate biomaterial for 3D cell culture. The organic material of meniscus mainly consists of collagen, especially collagen type I (Col I). Thus, Col I gel as natural biomaterial has been used for meniscus tissue engineering. As the mechanical properties of gels are weak and the meniscus is wedge-shaped, mechanical compression with simultaneous shaping has been applied to densify and form collagen.

Wedge-shape compression in a customized bioreactor system with a movable wedge was successfully established. By the compression to a defined volume the compression factor was determined. As meniscal cells are a limited cell source and have limited proliferation potential, mesenchymal stem cells (MSC) – providing fibrochondrogenic potential were directly isolated from human bone marrow and their chondrogenic potential was confirmed by histological stainings.

Human MSC were embedded in Col I gel. MSC-laden Col I gels were compressed to a wedge-shape 3D meniscus model. During three weeks of static 3D culture, wedge-shaped MSC-laden Col I gels were analyzed in their shrinkage behaviour. After three weeks, wedge-shaped MSC-laden Col I gels showed high viability and were investigated by immunohistochemical analysis and by quantitative GAG/DNA assays.

The here reported study suggested a wedge-shaped MSC-laden Col I gel as suitable biomaterial for meniscus tissue engineering. The combination of the wedge-shape MSC-laden Col I gel with a 3D biological vascularized scaffold (BioVaSc®) – providing a vascularized structure after seeding with endothelial cells – is planned to build up a 3D vascularized meniscus model

Development of Bioactive Bioglass scaffolds coated with iron-loaded hydroxyapatite nanocomposites as potential biomaterials for bone tissue repair

Maria Laura Dittler[1], Mónica C. Gonzalez[2], Aldo R. Boccaccini[1]

[1] *Institute of Biomaterials, Department of Materials Science and Engineering*

University of Erlangen Nuremberg Cauerstr. 6, 91058 Erlangen, Germany

[2] *INIFTA-CCT CONICET (La Plata), National University of La Plata,*

Diag. 113 y 64 cc. 16 suc. 4 (1900) La Plata, Argentina

The major calcium phosphate (CaP) component of bone is not a chemically homogenous material. In vivo bone apatite efficiently exchange Ca^{2+} and PO_4^{3-} ions with CO_3^{2-}, F^-, Mg^{2+}, Na^+, and K^+ ions present in physiological fluids, which are believed to be one of the reasons for its biological response. In literature is well known that ion substitution strategies improve the physicochemical and biological response of synthetic hydroxyapatite. Lately, the design of bioactive glasses scaffolds has also gained much interest in tissue engineering. The goal of this study is to obtain Bioglass 45S5® Scaffolds (BGS) coated with magnetic Fe-containing hydroxyapatite nanoparticles (Fe-nHAp).

BGS were obtained by the foam replica technique [1]. Formation of Fe-nHAp surface films were produced by dip-coating for ten minutes in a 5 mg/ml Fe-nHAp suspension. The presence of iron on the surface was confirmed by EDX, magnetic susceptibility and by MP-AES. The microstructure and porosity of the scaffolds were observed by SEM and surface functional groups characterized by FTIR. Mineralization assays were performed to evaluate the material bioactivity.

The obtained evidence indicates that BGS were effectively coated with Fe-nHAp. Immersion of Fe-nHAp-covered BGS in simulated body fluid (SBF) lead to the formation of CaP cauliflower structures observed by SEM and similar to those formed by bare BGS under identical conditions, thus sustaining the bioactivity of the novel Fe-nHAp-covered BGS.

[1] Chen QZ, Thompson ID, Boccaccini AR. Biomaterials 27 (2006), 2414–25.

Development and characterization of hybrid hydrogel based on alginate di-aldehyde, gelatin and silk fibroin for tissue engineering applications

Supachai Reakasame and Aldo R. Boccaccini

Institute of Biomaterials, Department of Materials Science and Engineering

University of Erlangen-Nuremberg, Cauerstrasse 6, 91058 Erlangen, Germany

Silk fibroin (SF) has been widely used in tissue engineering because it is well known for the biocompatibility, biodegradability and mechanical properties. However, SF exhibits poor cell adhesion because of its hydrophobicity1. Gelatin is the material of choice used to promote cellmaterial interaction because it contains RGD (arginine-glycine-aspartic acid), cell adhesive peptide sequences. In our previous work, it was found that alginate di-aldehyde (ADA) could covalently crosslink with gelatin resulting in a slower gelatin release rate from ADA-gelatin hydrogels compared to that of alginate-gelatin blend hydrogels2. The present study is aimed to prepare and to assess the properties of a novel type of hybrid hydrogel based on ADA, gelatin and SF. ADA was synthesized via periodate oxidation of alginate. SF was extracted from Bombyx mori cocoons by the treatment with sodium carbonate and lithium bromide, respectively.

The hybrid hydrogels were prepared using a sonication technique. The weight loss and FTIR of hydrogels were investigated. The microstructure of the hydrogel film was investigated by scanning electron microscopy (SEM). The results demonstrated that the composition of the hydrogels had a significant effect on their physical properties. Mouse embryonic fibroblasts (MEFs) were grown on the hydrogels. Cell viability on the different hydrogels was compared by WST-8 assay. The results indicated that the ADA/gelatin/SF hydrogels at proper ratio supported cell attachment and proliferation. The results proved that such novel hybrid hydrogels might be tuned to obtain materials suitable for tissue engineering applications.

References

1. Nasim Annabi, Ali Tamayol, Jorge Alfredo Uquillas, Mohsen Akbari, Luiz E. Bertassoni, Chaenyung Cha, Gulden Camci-Unal, Mehmet R. Dokmeci, Nicholas A. Peppas, and Ali Khademhosseini. "An axial distribution of seeding, proliferation, and osteogenic differentiation of MC3T3-E1 cells and rat bone marrow-derived mesenchymal stem cells across a 3D Thai silk fibroin/gelatin/hydroxyapatite scaffold in a perfusion bioreactor" Materials Science andEngineering C 2016, 58: 960–970.

2. B. Sarker et al. "Alginate-based hydrogels with improved adhesive properties for cell encapsulation" International Journal of Biological Macromolecules 2015, 78: 72-78.

Functional polyelectrolyte coatings in prevention of medical devices from bacterial colonization

Aldona Mzyk[1], Justyna Straub[1], Martijn Riool[2], Leonie de Boer[2], Roman Major[1], Sebastian Jan Zaat[2]

[1] *Institute of Metallurgy and Material Science, Polish Academy of Sciences, Krakow, Poland*

[2] *Academic Medical Center/University in Amsterdam, Amsterdam, The Netherlands*

Essential strategies to prevent biomaterial associated infections are based on preventing bacterial adhesion on the surface of biomedical devices or providing contact/release killing activity. The Polyelectrolyte Multilayer Films (PEMs) made of natural polymers are one of the approaches to obtain extracellular matrix (ECM) mimicking coatings which may prevent implant colonization by microorganisms, facilitate its surface repopulation by the blood circulating endothelial progenitor cells, mask the material from an inflammatory response, and reduce risk of thrombosis. The physico-chemical properties of multilayer coatings may be easily modulated, therefore PEMs are a suitable system to investigate microbial response depending on surface characteristics. Among natural polymers, the PEM systems composed of chitosan/chondroitin sulphate and albumin/fucoidan were chosen as the most promising blood contacting materials due to efficient endothelialization and hemocompatibility.

The polymer coatings have been investigated in terms of chemical structure, surface morphology, wettability, stiffness as well as human cells response. The properties of the films were regulated by chemical cross-linking or nanoparticles incorporation. In this studies an effect of the commonly described N-hydroxysulphosuccinimide/1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (NHS/EDC) cross-linking chemistry were compared with results of PEMs modification by an alternative, less cytotoxic cross-linking agent genipin. Two different types of nanoparticles, i.e. silver (Ag) or reduced graphene oxide (rGO) have been taken into consideration due to increasing interest in their potential broad antimicrobial activity. Selected coatings have been subjected to the contact/release antimicrobial activity tests against Gram-positive (S. epidermidis, S. aureus) and Gram-negative (E. coli) strains. The JIS, MIC/MBC and agar diffusion assay was performed. Significant contact/release antimicrobial activity has been found for the Chi/CS and ALB/FU coatings with incorporated AgNPs, as well as ALB/FU cross-linked by genipin. Surfaces containing graphene oxide flakes did not show any antimicrobial activity.

Acknowledgements: The research was financially supported by the COST Action TD1305 and statutory funding of the IMM PAS in Poland (Z-2) as well as the AMC in the Netherlands.

Electrophoretic deposition of gentamicin loaded chitosan/gelatin/bioactive glass composite coatings on PEEK/bioactive glass layers: A comprehensive study on in-vitro-bioactivity and antibacterial effect

M. Atiq Ur Rehman[1]*, Wolfgang H. Goldmann[2], Aldo R. Boccaccini[1]

[1] *Institute of Biomaterials, Department of Material Science and Engineering, University of Erlangen-Nuremberg, Cauerstr.6, 91058 Erlangen, Germany*

[2] *Department of Biophysics, University Erlangen-Nuremberg, Henkstr.91, 91052 Erlangen, Germany*

**muhammad.rehman.ur@fau.de*

This study presents the electrophoretic deposition (EPD) of gentamicin loaded in/bioactive glass (BG) coatings on PEEK/BG layers (deposited by EPD on 316L stainless steel). Initially, chitosan/gelatin/BG coatings were optimized on stainless steel by using design of experiment (DoE) approach. The 'best' results were obtained at the deposition voltage of 30 V, deposition time of 5 minutes and inter-electrode distance of 1 cm. The optimized concertation ratio between chitosan and gelatin was 1. Once the EPD parameters were optimized for stainless steel, similar suspension compositions and EPD parameters were employed on PEEK/BG layers. However, due to the insulation effect offered by the PEEK/BG layer deposition voltage was increased to 50 V in order to form fairly homogenous coatings.

The resulting composition, microstructure, in-vitro bioactivity and antibacterial activity were investigated. The presence of chitosan/gelatin/BG in the coatings was confirmed by scanning electron microscopy (SEM) and Fourier transform infrared spectroscopy (FTIR). The presence of gentamicin sulphate in the coatings was verified by electron dispersive spectroscopy (EDX) and ultraviolet/visible (uv/vis) spectroscopy. The developed coatings form hydroxyapatite layer upon immersion in SBF for three days. Moreover, the chitosan/gelatin matrix shows the desired degradation behaviour upon immersion in SBF. The gentamicin was released in the sustained manner; and minimum inhibitory concentration was maintained even after three weeks. Furthermore, the gentamicin provided antibacterial effect against staphylococcus carnosus and Escherichia coli.

Tribocorrosion response of different multilayer TaN coatings in biological environments

R. Bayón, V. Sáenz de Viteri, L. Mendizabal, J. Barriga, A. Igartua

IK4-Tekniker Research Alliance
Eibar, Spain

The principal cause of failure of biomedical implants is due to the tribological processes (friction and wear) they are exposed to during their lifetime. Besides, these processes are found to accelerate the corrosion mechanisms occurring in the aggressive environment the body represents. The first contacting area is the surface; therefore, the problem can be minimized by the development and application of adequate coatings combining both low friction and high corrosion and wear resistance.

Tantalum nitride (TaN) is a hard, chemically inert, highly corrosion resistant biocompatible material. These features make it an excellent candidate for application as protective coating on biological corrosive environments. Recently developed high power pulsed magnetron sputtering (HPPMS) is a physical vapor deposition technology in which the power is applied to the target in pulses of low duty cycle and frequencies leading to pulse target power densities of several kW cm-2. This mode of operation results in a dense plasma discharges with high degree of ionization of sputtered particles. This fact fosters the deposition of columnar-free, fully dense and less defective TaNx coatings previously infeasible by conventional dc magnetron sputtering (dcMS). The multilayer structure is created by alternately switching two different pulse shapes and maintaining each pulse shape for a specific pulse repeat duration on the same target during a single process.

In the present work, multilayer TaN coatings characterized by different bilayer periods have been deposited on titanium biomedical alloys. Corrosion, wear and tribocorrosion performance has been analyzed in simulated body fluids with protein content. TaN films strongly decrease wear rates of titanium substrates when they are simultaneously submitted to electrochemical and tribological effects, being very attractive candidates for their use in biomedical field.

Pure titanium with enhanced properties applied to newly developed dental implant

K. Sztwiertnia[1], J. Kawalko[2], M. Bieda[1]*, D.Wojtas[1], K. Wierzbanowski[2],

M. Wronski[2], W. Pachla[3], M. Kulczyk[3]

[1] *Institute of Metallurgy and Materials Science, Polish Academy of Sciences, Krakow, Poland*

[2] *AGH University of Science and Technology, Krakow, Poland*

[c] *Institute of High Pressure Physics, Polish Academy of Sciences, Warsaw, Poland*

**m.bieda@imim.pl*

Commercial pure titanium (CP Ti) is the basic material applied for dental implants. Lately proposed prosthetic structure [1] due to its multifunctionality, have to carry high loads. Despite insufficient mechanical properties CP Ti is the most suitable material for this purpose because of good biocompability and corrosion resistance. The aim of the work was to improve the mechanical properties of CP Ti on the level comparable to that of Ti6Al4V, which can be achieved by eg. grain refinement. Titanium with nano- or ultra-fine crystalline microstructure can be obtained by various methods of Severe Plastic Deformation (SPD). The microstructure and mechanical properties of CP Ti grade 2 and grade 4 after three processes - ECAP, hydrostatic (HE) and KoBo extrusion - were compared. Initial titanium samples were characterized by a typical coarse microstructure with average grains diameter about 50 μm. All methods: ECAP, HE and KoBo allowed to obtain significant strengthening. The highest grain refinement capability was observed after HE, while KoBo and ECAP both yield a smaller fine-grained fraction. This corresponded to improved mechanical properties. Yield strength reached 950 MPa after HE, while 450 MPa after KoBo, and 690 MPa after ECAP.

[1] Z. Machynia, K. Koynov, K. Sztwiertnia, Dental Distraction Implant, Patent No. P.416 895.

Acknowledgement

This work has been supported by the National Centre for Research and Development in Poland through the Project No PBS2/A6/18/2014.

Development of a conductive, biomimetic, polymeric fiber mat for a bilayered cardiac patch

Lena Vogt, Florian Ruther

Lehrstuhl für Biomaterialien, Friedrich-Alexander Universität Erlangen-Nürnberg
91058 Erlangen, Germany

In contrast to other tissues such as bone or skin, myocardial tissue exhibits very limited intrinsic regeneration ability. Due to this shortage, an event like myocardial infarction could lead to the loss of cardiac cells and the formation of scar tissue, eventually resulting in chronic or congestive heart failure. Therefore, therapeutic strategies to repair the damaged tissue, e.g. the cardiac patch strategy, become increasingly attractive [1].
In this study a novel type of conductive, biomimetic polymer fiber mat suitable for cardiac tissue engineering applications will be fabricated and characterized. The electrospun fiber mat, which will serve as a top layer of a bilayered cardiac patch, will be based on synthetic, electrically conductive polymers as well as natural components of the extra cellular matrix. The choice of fabrication technique and materials should provide electrical conductivity, matching mechanical and degradation properties as well as an anisotropic structure similar to native tissue to provide cell contact guidance.

[1] H. Jawad, et al., Myocardial tissue engineering: a review, J. Tissue Eng. Regen. Med. 1 (2007) 327–342

Cinnamon bark oil encapsulated poly (ε-caprolactone) nanofiber mats for wound healing

Irem Unalan, Aldo R. Boccaccini

Institute of Biomaterials, University of Erlangen-Nuremberg
91058 Erlangen, Germany

Essential Oils (EOs) based on plant extract, such as cinnamon bark oil, have been used to accelerate the wound healing process since ancient times due to various therapeutic benefits such as antibacterial and antimicrobial activities, inflammatory and antioxidant potential [1,2]. However, these EOs have poor physical properties such as high sensitivity to degradation, and volatility, making them difficult to be used in wound dressing applications. Electrospinning is a convenient way to produce fibrous structures with enhanced releasing capability and able to exploit the therapeutic properties of EOs[3,4]. The objective of this study is to produce cinnamon bark oil (CIN) encapsulated poly(ε-caprolactone) (PCL) nanofiber mats by electrospinning technique for wound healing applications. The surface morphology of the mats was characterized using scanning electron microscopy (SEM) and the average diameter of electrospun fibers was estimated from SEM micrographs via Image J program. The average fiber diameter for the pure PCL fiber mat was measured to be 99± 15 nm, which increased to 110± 13 nm upon incorporation of CIN. The functional groups present in the mats were analyzed using Fourier transform infrared (FTIR) spectroscopy to determine the presence of CIN on the surface of CIN/PCL nanofiber mats by investigating aldehyde and carbonyl group (C=O) in the IR spectrum. The wettability of the pure PCL and CIN/PCL nanofiber mats were measured using a contact angle meter with the sessile drop method and results showed that the wettability of the mats is increased with increasing concentration of CIN.

[1] S. Prabuseenivasan, M. Jayakumar, S. Ignacimuthu, In vitro antibacterial activity of some plant essential oils, BMC Complement Altern Med. 6 (2006) 39. doi:1472-6882-6-39 [pii]\r10.1186/1472-6882-6-39.

[2] G. Jin, M.P. Prabhakaran, D. Kai, S.K. Annamalai, K.D. Arunachalam, S. Ramakrishna, Tissue engineered plant extracts as nanofibrous wound dressing, Biomaterials. 34 (2013) 724–734. doi:10.1016/j.biomaterials.2012.10.026.

[3] P. Wen, D.-H. Zhu, H. Wu, M.-H. Zong, Y.-R. Jing, S.-Y. Han, Encapsulation of cinnamon essential oil in electrospun nanofibrous film for active food packaging, Food Control. 59 (2016) 366–376. doi:10.1016/j.foodcont.2015.06.005.

[4] K.A. Rieger, J.D. Schiffman, Electrospinning an essential oil: Cinnamaldehyde enhances the antimicrobial efficacy of chitosan/poly(ethylene oxide) nanofibers, Carbohydr. Polym. 113 (2014) 561–568. doi:10.1016/j.carbpol.2014.06.075.

Synthesis, characterisation and applications of ordered mesoporous silver doped bioactive glass

Francesca Elisa Ciraldo, Aldo R. Boccaccini

Institute of Biomaterials, Department of Materials Science and Engineering
Friedrich-Alexander University Erlangen-Nuremberg, 91058 Erlangen, Germany

The demand for new engineered materials has increased in the last years. Bioactive glasses are commonly used in biomedical applications thanks to their ability to form a biologically equivalent apatite surface once in contact with biological fluids. Mesoporous bioactive glasses (MBGs) are usually based on the ternary system SiO_2-CaO-P_2O_5 and characterised by large specific surface area, highly tailored ordered porosity and pore size in the range of 5-20 nm. These features make them promising candidate to be used as local drug delivery system.

In this work, silver doped MBGs (Ag-MBGs) were synthesized employing the evaporation induced self-assembly (EISA) process and using Pluronic F127 as structure directing agent. The resulting material was characterised by using TEM and BET to evaluate the inner microstructure, the specific surface area and the pore volume distribution. Bioactivity tests in simulated body fluid (SBF) were performed to assess the ability of the Ag-MBG to develop hydroxyl-carbonate-apatite. The versatility of Ag-MBGs was also demonstrated by using the MBG particles to coat 3D scaffold for bone tissue engineering applications.

Electrophoretic deposition of zein/bioactive glass composites

Laura Ramos-Rivera[1], Nima Meyer[1], Luca Tortoreto[2], Aldo R. Boccaccini[1]

[1] *Institute of Biomaterials, Department of Materials Science and Engineering*
University of Erlangen-Nuremberg, 91058 Erlangen, Germany

[2] *Rina Consulting - Centro Sviluppo Materiali SpA*
Business Unit: Materials, Technology & Innovation, Via di Castel Romano – Rome, Italy

Electrophoretic deposition (EPD) has been used to fabricate composite coatings based on the natural polymer zein combined with bioactive glass (BG) particles on stainless steel substrates. Through the deposition of various BG compositions, namely 45S5 BG and Cu-doped BG, this work sought to demonstrate the ability of the films to potentiate the formation of hydroxyapatite (HA). The experimental conditions towards homogeneous surface and good distribution of bioglass particles are described. The formation of HA at the surface of the coatings following immersion in simulated body fluid (SBF) was confirmed using Fourier transform infrared spectroscopy (FTIR). Tribological studies show high adhesion of the coating with the substrate in some of the conditions and low hardness.

Biodegradable zinc alloy with magnesium subjected to hydrostatic extrusion: Evolution of microstructure, mechanical properties and corrosion behavior

A. Jarzębska[1]*, M. Bieda[1], Ł. Rogal[1], R. Chulist[1], J. Guśpiel[1], M. Strąg[1], K. Sztwiertania[1] W. Pachla[2], M. Kulczyk[2]

[1] *Institute of Metallurgy and Materials Science, Polish Academy of Sciences, Krakow, Poland*

[2] *Institute of High Pressure Physics, Polish Academy of Sciences, Warsaw, Poland*

* *a.jarzebska@imim.pl*

New application of zinc was recently found out as a biodegradable metal suitable for use in implants e.g. stents. Previous investigations revealed that zinc possesses excellent degradation rate and acceptable cytocompatibility. However, the drawback of zinc are its insufficient mechanical properties. In order to obtain better mechanical properties pure and low-alloyed zinc were intensively studied in past few years.

Unconventional method of extrusion called hydrostatic extrusion (HE) was proposed to low-alloyed zinc in order to enhanced mechanical properties. Zinc alloy with 1 % wt. of magnesium was prepared by gravity casting and then subjected to plastic deformation by HE. Microstructural, mechanical and corrosion investigations were then carried out.

Longitudinal cross section to extrusion direction was characterized using Electron Backscatter Diffraction technique performed on FEI Quanta 3D FEGSEM. Mechanical properties were specified based on tensile test and microhardness measurements. Information about corrosion behavior was gained from both potentiodynamic and immersion tests.

Obtained results revealed that HE positively influenced grain refinement. Grains with average size 1 μm were observed. Such reduction in grain size provoked improvement of mechanical properties. YS = 316 MPa, UTS = 435 MPa and E = 35 % were achieved. Grain refinement also influenced corrosive properties. Based on potentiodynamic curves corrosion rate equal 10 μm/year was calculated. Those properties meet requirements for stents applications and it seems that low-alloyed zinc is suitable for such use. Further biocompatibility investigations of material processed in that way are essential for stents developments.

Preparation and characterization of mesoporous calcium doped silica coated TiO2 scaffolds and their drug releasing behaviour

Abirami Sengottuvelan

Institute of Biomaterials, Friedrich-Alexander-University Erlangen-Nuremberg,Germany

Three dimensional titanium dioxide (TiO_2) scaffolds are receiving attention for the reconstruction of damaged bone tissue. Additional to their biocompatibility, these scaffolds show mechanical properties similar to bone and therefore meet the requirements for the usage as a scaffold system. However the bioactivity of these scaffolds is comparatively lower than that of bioactive glass scaffolds [1]. Sol-gel derived mesoporous bioactive glasses exhibit high bioactivity in vitro and can also be loaded with drugs due to its tunable pore size [2]. Hence, the main aim of this work is to produce and characterize TiO2 scaffolds coated with sol-gel derived mesoporous calcium silicates (MCS) and to test their in vitro bioactivity and drug delivery properties. TiO2 scaffolds were coated with different wt% of MCS using a simple dip coating method. The influence of the coatings on the bioactivity, e.g. hydroxyapatite formation, was measured in vitro in simulated body fluid (SBF). The scaffolds coated with 4 wt% of MCS showed high bioactivity after one week of immersion in SBF. The formation of bioactivity was confirmed by SEM, EDS and XRD. Ibuprofen drug storage capabilities and release rate should be investigated in MCS and MCS coated TiO2 scaffolds.

[1] Cachinho, S. C. P. & Correia, R. N. Titanium scaffolds for osteointegration: mechanical, in vitro and corrosion behaviour. J. Mater. Sci. Mater. Med. 19, 451–7 (2008).

[2] Li et al., A template route to the preparation of mesoporous amorphous calcium silicate with high in vitro bone-forming bioactivity, J. Biomedical Materials Research Part B: Applied Biomaterials, 83B, 431–439 (2007).

Applications of low energy electron beam technology for sterilization and surface modification of medical products

Gaby Gotzmann

Fraunhofer-Institut für Organische Elektronik, Elektronenstrahl- und Plasmatechnik FEP
Dresden, Germany

Many new medical products cannot be sterilized by standard techniques like hot steam or exposure to sterilizing gases, as they are temperature sensitive, contain electronic parts like microchips or consist of polymeric materials. Also, the use of gamma irradiation for such applications is problematic due to long exposure time under radical atmosphere which leads to an increased degradation and therefore to a loss of functionality.

Using low energy electron irradiation it is possible to sterilize medical surfaces within some seconds because of very high dose rates. Therefore, degradation processes can be significantly reduced compared to gamma irradiation. In addition, it is possible to define the penetration depth of the electron beam into the product in order to prevent electronic parts from damage. Therewith, also highly sensitive objects like flexible OLEDs or bi-directional sensor-display assemblys can be sterilized safely and used for highly functionalized medical applications. Besides sterilization of such senstitive products, it is also possible to use low energy electron beam irradiation for modification and sterilization of diverse biological materials like peptides, collagen matrices, transplantation tissues or hydrogels. The surface modification by low energy electron beam can be used to adapt surface characteristics like hydrophilicity and create bacteria repellent surfaces or distinct growth patterns for human cells.

The Fraunhofer FEP focuses on the adaption of the low energy electron beam processes for new applications, but also to develop inline-capable systems for sterilization and modification applications in production processes and as batch systems. Besides, FEP researchers develop flexible OLEDs and miniaturized sensor and display structures, which also can be used for medical applications.

The effect of chemical composition on viscoelastic properties of methylcellulose/agarose hydrogel

B. Niemczyk*, P. Sajkiewicz

Institute of Fundamental Technological Research, Polish Academy of Sciences
Pawińskiego 5b, 02-106 Warsaw, Poland

** bniem@ippt.pan.pl*

Appropriate kinetics of hydrogels cross-linking is very important, particularly for thermosensitive materials directed toward tissue engineering applications. In this regard methylcellulose is widely used, however there is only a handful of publications describing the characteristics of the methylcellulose/agarose composite. The aim of the studies is to analyse the kinetics of cross-linking of aqueous solutions of methylcellulose and methylcellulose/agarose using dynamic mechanical analysis (DMA).

The oscillatory analysis was performed for few concentrations of pure methylcellulose and methylcellulose/agarose utilizing small-amplitude sinusoidal deformation. In the temperature range 33-39°C, under isothermal conditions time dependence of storage G', viscous modulus G'' and complex viscosity were determined in limited range of time. Considering that there is no intersection of G' and G'' curves, the kinetics of cross-linking was deduced from the time derivative of the storage modulus G'. As a parameters of cross-linking, the time position and the height of the maximum of the time derivative of the storage modulus were taken. The numerical analysis including approximation and extrapolation beyond the registered time with asymmetric double sigmoidal function as well as integration allows estimation of the final modulus and complex viscosity of hydrogels which is crucial from the practical perspective.

Our results show that addition of agarose to methylcellulose hydrogel at the w/w ratio 1:2 and 1:3 does not affect very much viscosity what is important for injectability. However at physiological temperature the presence of agarose influences the cross-linking kinetics leading to higher rate of cross-linking (higher maximum of the storage modulus derivative) and the final value of the storage modulus. For instance, at 35°C agarose addition leads to an increase of the final G' and complex viscosity 1100- and 13-times respectively compared to pure methylcellulose. Moreover, addition of agarose results in increase of the time position of the maximum of crosslinking rate.

Keywords: methylcellulose, agarose, hydrogel, cross-linking kinetics, DMA, modulus

Magneto-Plasmonic nanoparticles for photothermal therapy

C. Multari[1], M. Miola[1], F. Laviano[1], R. Gerbaldo[1], G. Pezzotti[2], D. Debellis[3], E. Vernè[1]

[1] *Politecnico di Torino, Torino, Italy*

[2] *Kyoto Institute of Technology, Kyoto, Japan*

[3] *Italian Institute of Technology, Genova, Italy*

Nanoparticles have been widely studied for their ability to be used in cancer treatment. Particularly, magneto-plasmonic nanoplatforms (MPNPs), composed by superparamagnetic iron oxide nanoparticles (SPIONs) and gold nanoparticles (GNPs), are of great interest because they possess both magnetic and plasmonic properties.

A reproducible synthesis method is used to obtain MPNPs made of a magnetic core and an external gold decoration. SPIONs are prepared by co-precipitation method and GNPs are synthetized from soluble gold salts by reduction and then added to the SPIONs suspension, in order to promote their growth on the SPIONs surface.

The correct formation of MPNPs and their composition are detected by physical characterization that gives information about size and morphology. The superparamagnetic behavior of SPIONs and the plasmonic properties of GNPs are also verified thanks to magnetic and optical characterization respectively.

A cytotoxicity study is performed comparing healthy and cancer cells exposed to MPNPs. To detect the efficacy of GNPs, a green laser source is used in order to evaluate the ability of GNPs to convert absorbed light into thermal energy.

Cell tests confirm that MPNPs causes an important damage of cancer cells, if exposed to laser light; while is not resulting dangerous in normal cells. This indicate that the MPNPs allows to convert the light received into heating which can destroy cancer cells, due to their high heat sensitivity.

Using these MPNPs is possible to have a new approach to cancer therapy that consents to drive MPNPs directly in tumor site, to use them as drug delivery system and contrast agent and contemporaneously is possible to use MPNPs for photothermal therapy.

Evaluation of mechanical properties and biocompatibility of Gum Metal for implant applications

Karol M. Golasiński[1]*, Elżbieta Pieczyska[1], Rainer Detsch[2], Aldo R. Boccaccini[2], and Naohisa Takesue[3]

[1] *Institute of Fundamental Technological Research, Polish Academy of Sciences, Pawińskiego 5B, 02-106 Warsaw, Poland*

[2] *Institute of Biomaterials, Friedrich-Alexander University of Erlangen-Nürnberg, Cauerstraße 6, 91058 Erlangen, Germany*

[3] *Department of Applied Physics, Graduate School of Science, Fukuoka University, Nanakuma, Jonan-ku, Fukuoka 814-0180, Japan*

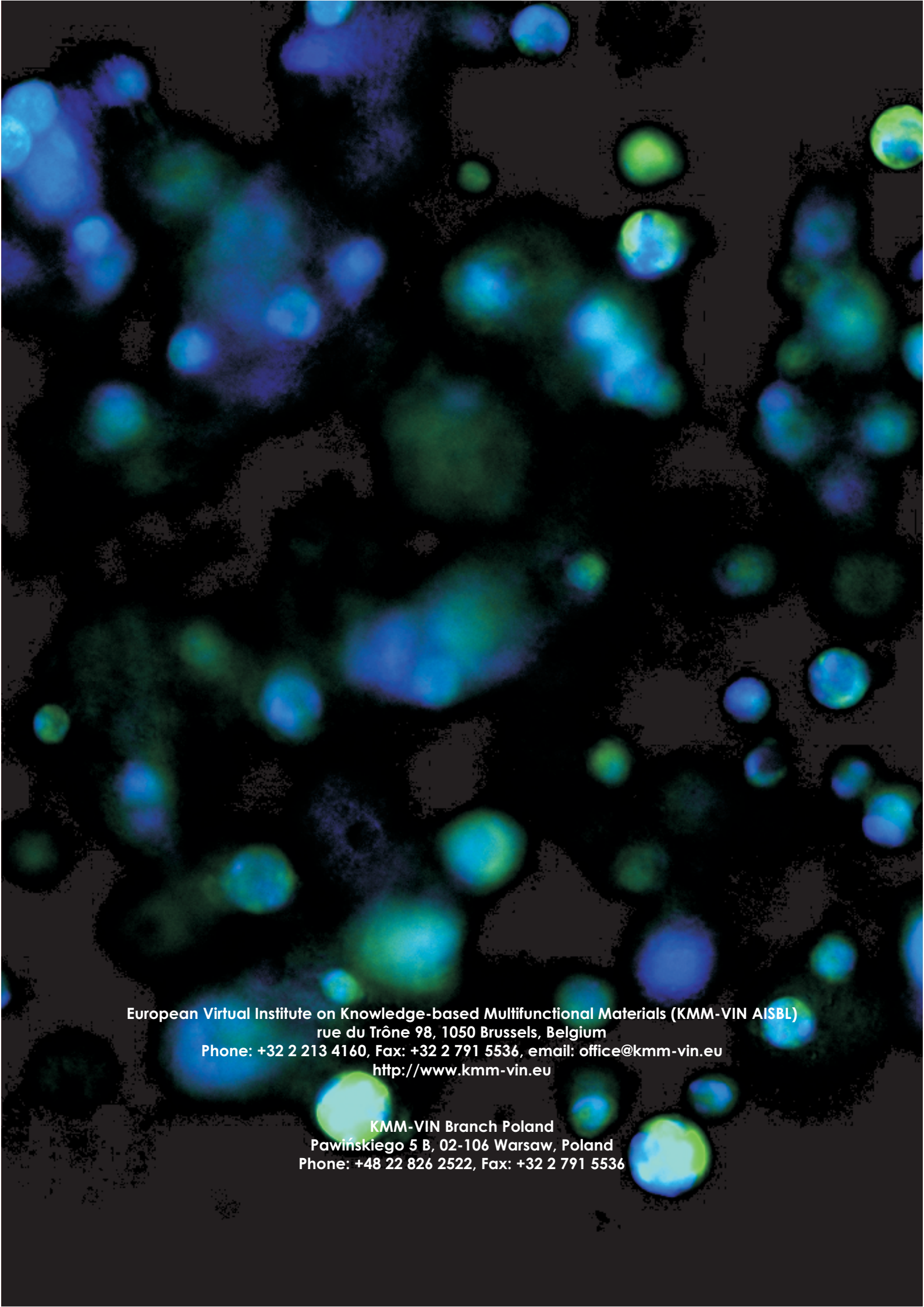
**kgolasin@ippt.pan.pl*

Titanium and titanium alloys are widely applied as materials for orthopedic and dental implants. However currently, a great limitation of these materials is their high Young's modulus (over 100 GPa) which often leads to bone-shielding effect. Another drawback of certain Ti-based alloys is a content of cytotoxic elements; for example of Al and V in Ti-6Al-4V (ELI). That is why, for the last decades a major effort has been put to develop a Ti-based material with lower Young's modulus and negligible toxicity.

In this work, mechanical properties of a β-Ti alloy Gum Metal (Ti–23Nb–0.7Ta–2.0Zr–1.2O at.%, free of cytotoxic content), which was fabricated at Toyota Central Research & Development Laboratories, Inc., were investigated. It was confirmed that Gum Metal is characterized by a low Young's modulus (around 60 GPa), high strength (over 1000 MPa) and a large range of reversible deformation, which are important features in the context of potential implant applications. Moreover, a comprehensive assessment of biocompatibility was realized. Properties of Gum Metal were contrasted with those of Ti-6Al-4V (ELI) which was taken as reference. Surface conditions, such as topography, roughness and structural composition, were analyzed. Evaluation of biocompatibility for the alloys was performed by cell attachment and spreading analysis after predefined cell culture periods. Gum Metal presented excellent properties, what makes it a good candidate for implant applications.

Acknowledgements

The research has been supported by the KMM-VIN Research Fellowship (Call 9) and the Polish National Science Centre (NCN) under Grants nos. 2014/13/B/ST8/04280 and 2016/23/N/ST8/03688.



European Virtual Institute on Knowledge-based Multifunctional Materials (KMM-VIN AISBL)
rue du Trône 98, 1050 Brussels, Belgium
Phone: +32 2 213 4160, Fax: +32 2 791 5536, email: office@kmm-vin.eu
http://www.kmm-vin.eu

KMM-VIN Branch Poland
Pawińskiego 5 B, 02-106 Warsaw, Poland
Phone: +48 22 826 2522, Fax: +32 2 791 5536