Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P001

Eigenschaftsverbesserung von Polylactiden durch Erhöhung der Kristallinität

*S. Rakus[1]
[1]Opti-Polymers GmbH, Produktionsleitung , Rudolstadt, Deutschland

Fragestellung

Die Eigenschaften von Polylactiden sollen durch Erhöhung der Kristallinität verbessert werden. Polylactide, umgangssprachlich auch Polymilchsäuren (kurz PLA, vom englischen Wort polylactic acid) genannt, sind synthetische Polymere, die zu den Polyestern zählen. Sie sind aus vielen, chemisch aneinander gebundenen Milchsäuremolekülen aufgebaut.

Methoden: Erhöhung der Kristallinität durch Nukleirungsmittel

Durch Nukleirungsmittel kann die Kristallinität erhöht werden. Es erhöht sich sowohl die Kristallisationstemperatur als auch der Kristallisationsgrad (Figure 1). Nukleirungsmittel haben folgende Vorteile: Schnelle Erzeugung von feinen Kristallen zur Verbesserung der Verarbeitungsfähigkeit, Verbesserung der Wärmebeständigkeit ,Verbesserung der elastischen und thermischen Eigenschaften, Erhöhung der Steifigkeit und Stoßfestigkeit, Reduzierung der Zykluszeiten durch Kühlzeitreduktion.

Ergebnisse: Nukleirungsmittel Opti NK-30 Bio

Von der Opti-Polymers GmbH wurde das Nukleirungsmittel Opti NK-30 Bio entwickelt.Es verbessert die mechanischen Eigenschaften von Biopolymeren auf Basis PLA-Lignin. Es ermöglicht weiße und zylindrische Stranggranulate. Der Wirkstoff ist ein Blend aus feinteiligen anorganischen Produkten (Wirkstoffgehalt 30%). Das Produkt kann in Heißkanalwerkzeugen verarbeitet werden.

Die Dosierungsempfehlung beträgt 3 oder 6%.

Dosierung:	Zugabe von 3%	Zugabe von 6%
Kerbschlagzähigkeit [kJ/m2]	5	6
Zug-E-Modul [MPa]	2550	2600
Zugfestigkeit [MPa]	50	48
Bruchdehnung [%]	7	8

Schlussfolgerungen:

Opti NK-30 Bio hat folgende verarbeitungstechnische Vorteile:

Verbesserung der physikalischen Eigenschaften bei gespritzten Teilen (Figure 2 und 3),Verbesserung der Entformungseigenschaften ,Höhere Dimensionsstabilität der Bauteile (Verringerung des Verzuges) ,Produktkonstanz und gleichmäßige Formfüllung ,Verbesserung des Einzugsverhaltens der Biopolymere Reduzierung der Zykluszeiten durch Kühlzeitreduktion (Figure 4)

Figure 1

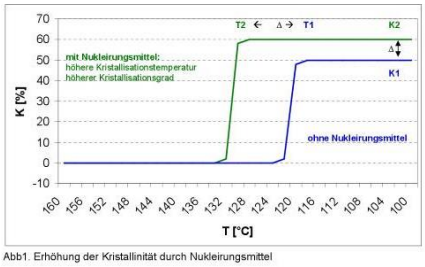
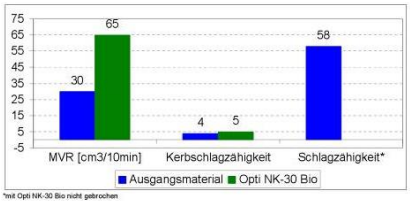


Abb1. Erhöhung der Kristallinität durch Nukleirungsmittel



*mit Opti NK-30 Bio nicht gebrochen

Abb.2: Verbesserung der physikalischen Werte (MVR, Kerbschlagzähigkeit, Schlagzähigkeit)

Figure 2

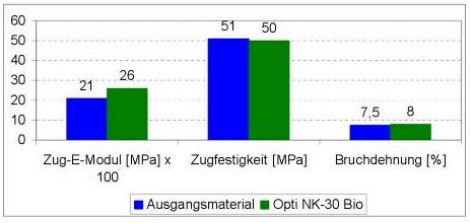


Abb.3: Verbesserung der physikalischen Werte (Zug-E-Modul, Zugfestigkeit, Bruchdehnung)

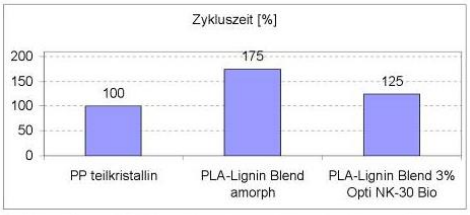


Abb.4: Reduzierung der Zykluszeiten bei der Herstellung eines 4 mm Zugstabes

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P002

Tailored Poly(glycidol) based Hydrogels for 3D-Bioprinting

*W. Smolan[1], T. Jüngst[1], J. Groll[1]
[1]University Hospital and Julius-Maximilians-University Würzburg, Department for Functional Materials in Medicine and Dentistry and Bavarian Polymer Institute , Würzburg, Deutschland

Poly(glycidol) (PG) is a potential alternative to the well-established poly(ethylene glycol) (PEG) for biomedical applications with similar biocompatibility but additional opportunity for side-chain functionalisation. PG can be synthesised with controlled chain length and low dispersity *via* living polymerisation techniques. The copolymerisation of different monomers enables to predefine the amount of functional groups which can be used for polymer analogue functionalisation, e.g. with cysteine groups for chemically orthogonal native chemical ligation (NCL) to form polymer-peptide hybrid molecules [1].

These tailored polymers and functionalisation techniques are enriching for biofabrication which is a fast developing research field with the need of new printable materials [2]. We have developed chemical crosslinked PG based hydrogels for 3D-printing mechanically stabilised *via* UV-light induced thiol-ene click chemistry [3]. Further, we have shown that recombinant spider silk protein possessing shear thinning properties due to *β*-sheet interactions, allows for 3D-printing of cell-loaded spider silk constructs *via* robotic dispensing without further crosslinking for mechanical stabilisation [4]. Inspired by this result, supramolecular interaction moieties for PG have to be identified that are needed to generate a printable system based on the side chain functionalised PG.

Here we present the linear statistical copolymerisation of ethoxy ethyl glycidyl ether (EEGE) with allyl glycidyl ether (AGE) in a ratio of EEGE:AGE = 54:6. After deprotection, the polymer analogue functionalisation *via* thiol-ene click chemistry with several functional groups, such as pyrene/naphthalene diimide and phosphonate/imidazole, were investigated. A study of the supramolecular hydrogel formation driven by *π-π* stacking and ionic interactions was performed. Finally, the gels" physico-chemical properties will be assessed with respect to 3D-printability.

References

[1] M. Kuhlmann, O. Reimann, C.P.R. Hackenberger, J. Groll, *Macromolecular Rapid Communications* **2015**, *36*, 472-476.
[2] T. Jüngst, W. Smolan, K. Schacht, T. Scheibel, J. Groll, *Chemical Reviews* **2016**, *116*, 1496-1539.
[3] S. Stichler, T. Jüngst, M. Schamel, I. Zilkowski, M. Kuhlmann, T. Böck, T. Blunk, J. Teßmar, J. Groll, *Annals of Biomedical Engineering* **2016**, *45*, 273-285.
[4] K. Schacht, T. Jüngst, M. Schweinlin, A. Ewald, J. Groll, T. Scheibel, *Angewandte Chemie International Edition* **2015**, *54*, 2816-2820.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P003

Cross-linked Hyaluronic Acid Hydrogels for 3D Printing

*J. Shan[1], M. Weis[1], E. Wirth[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, Abteilung für Funktionswerkstoffe der Medizin und Zahnheilkunde , Würzburg, Deutschland

Introduction

Due to fast development of biofabrication, printable hydrogels have become promising materials as bioinks for 3D printers in medical applications. Therefore, rheological and mechanical issues during and after printing process as well as swelling behavior, cytocompatibility and biodegradation must be considered when designing new hydrogels[1]. Hyaluronic acid (HA) as natural biopolymer already fulfill biological prerequisites. Aldehydes as functional groups in HA chains enable formation of hydrogels via cross-linking with dihydrazides or polyamines. This work focuses on synthesis of aldehyde modified HA derivatives and cross-linking reactions for hydrogel formation as well as the investigation of rheological and mechanical properties and cytocompatibility of the hydrogels.

Methods

HA was dissolved in water (1 % (w/v)) and sodium periodate ($NaIO_4$) solution was added. The molar ratio of $NaIO_4$ to HA varied between 0.5:1, 1:1, 2:1 and 4:1. After 2 h the reaction was stopped by an excess amount of ethylene glycol. The mixture was dialyzed against H_2O (MWCO = 3500 Da) for 72 h and lyophilized. The formation of hydrogels was performed with various amounts of oxidized HA, adipic acid dihydrazide (ADH) and gelatin in PBS. Rheological measurements for shear thinning tests were executed with a cone-plate setting and shear rates between 0.01 s^{-1} and 2000 s^{-1}. Mechanical tests were performed at a Universal Testing Machine with a 100 N load cell and maximum penetration depth of 2 mm. The pressure of the 3D printer was 2.0 bar and the gel was pressed through a cone with a diameter of 0.25 mm.

Results and Conclusions

Rheological and mechanical issues of the hydrogels depended on the experiential oxidation degree of HA, the ratio of cross-linkers to HA and the method of sample preparation. In general, HA with higher experiential oxidation degree led to decreasing viscosity and mechanical firmer hydrogels. Gels prepared with vortex mixer were also more stable than shaken and heated samples. Shear thinning behavior of the gels was proved by rheological measurements.

3D printing showed that the gel with 3.50 wt.-% high-molecular HA, oxidized with the molar ratio of 1:2 to $NaIO_4$, and 0.075 wt.-% ADH provided the best results.

References

[1] T. Jungst, W. Smolan, K. Schacht, T. Scheibel, J. Groll, *Chem. Rev.* **2016**, *116*, 1496-1539.

Figure 1

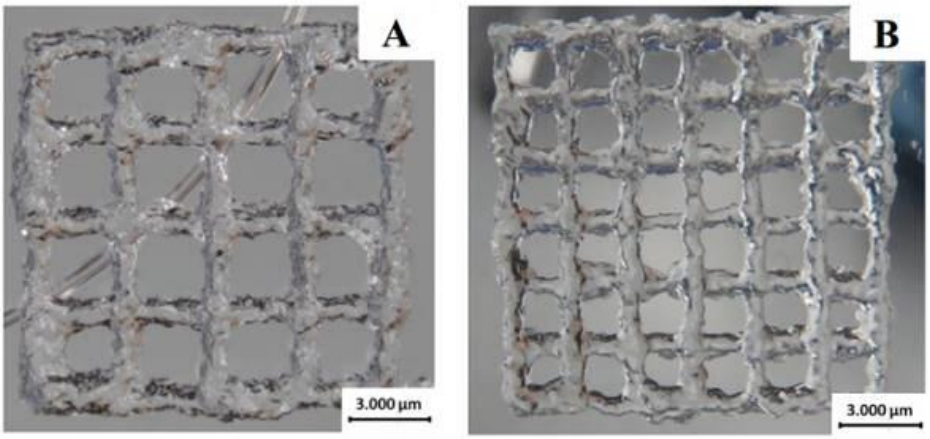


Figure 1 Printing with mixture 3.50 wt.-% oxidized HA and 0.075 wt-% ADH, 8 layers; **A:** 2 mm between each strand; **B:** 3 mm between each strand

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P004

Drop-on-demand bioprinting of a vascularized liver lobule-on-a-chip model

*A. Blaeser[1], M. Frissen[2], J. Schöneberg[1], M. Köpf[1], D. F. Duarte Campos[1], C. Trautwein[2], H. Fischer[1]
[1]RWTH Aachen University Hospital, Department of Dental Materials and Biomaterials Research , Aachen, Deutschland
[2]RWTH Aachen University Hospital, Clinic for Gastroenterology, Metabolic Disorders and Internal Intensive Medicine (Medical Clinic III) , Aachen, Deutschland

Background

Bioprinting offers huge potential for the generation of 3D in-vitro models for drug and toxicity testing. Due to its important role in drug-metabolism and protein synthesis, liver mimicking tissue analogues are of special interest in this field of research.

Aim

Here, we present a novel bioprinting strategy to generate vascularized, hepatocyte models with structural similarity to native tissue. We hypothesize that capillary-like network formation improves the biofunctionality, protein synthesis and urea secretion in those 3D-printed models.

Methods

Using a custom-built, drop-on-demand bioprinter fibrin networks loaded with human umbilical vein endothelial cells (HUVECs) and human mesenchymal stem cells (hMSCs) were printed. The pores of the network were filled with HUH7 hepatocyte laden agarose dots. Post-printing cell viability, cell spreading, and capillary-like network formation were analyzed for up to two weeks using fluorescence microscopy (FDA/PI), immunofluorescence staining (CD31, DAPI), and two-photon laser scanning microscopy. Albumin and urea secretion were studied for up to 21 days using 10 different combinations of hydrogels and cells (hepatocyte monoculture vs. triculture). In addition, the impact of capillary-like network formation on albumin and urea secretion was studied as a function of the sample thickness (300 µm vs. 1,800 µm).

Results

The fabricated hepatic models comprised a fibrin network with adjustable line width of 150-600 µm filled with hepatocyte containing agarose domains measuring 500 µm in diameter. Printed cells maintained high viability (> 94 %) and proliferation potential. Following two weeks of culture pronounced capillary formation could be observed in all bioprinted samples. For thin hepatic models (300 µm) no significant differences in albumin and urea secretion was observed between the printed and non-printed control group. However, in thick hepatic models (1,800 µm) the printed capillary-network significantly improved urea secretion compared to the non-vascularized control group.

Conclusion

Viability, motility, and functionality of the three applied cell types were shown not to be affected by the printing procedure. Remarkably, in thick hepatic models integration of capillary-like networks was shown to improve the removal of metabolic products from encapsulated hepatocytes. We conclude that capillary integration is a key step in shifting bioprinted 3D-models towards biomimetic tissue analogues.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P005

Agarose-collagen hydrogel blends as suitable materials for the manufacturing of three-dimensionally printed tracheal organ models

*M. Köpf[1], F. Kreimendahl[1,2], D. F. Duarte Campos[1], A. Blaeser[1], S. Ossenbrink[2], L. Thiebes[2], S. Jockenhövel[2], H. Fischer[1]
[1]Universitätsklinikum Aachen, Lehr- und Forschungsgebiet Zahnärztliche Werkstoffkunde und Biomaterialforschung , Aachen, Deutschland
[2]Institut für Angewandte Medizintechnik (AME), Helmholtz Institut, NRW Schwerpunktprofessur für Biohybride und Medizinische Textilien , Aachen, Deutschland

In recent years, novel biofabrication technologies have enabled the rapid manufacture of hydrogel-cell suspensions into tissue-mimicking constructs. The development of novel materials for biofabrication still remains a challenge due to a gap between contradicting requirements such as three-dimensional printability and optimal cytocompatibility. Therefore, we hypothesize that hydrogel blends of agarose and type I collagen are suitable materials for manufacturing of tracheal organ models with regard to 3D printability as well as the formation of a capillary-like network and a functional pseudostratified epithelium.

Agarose and collagen as well as hydrogel blends of both components were characterized rheologically by measuring the gelation temperature, gelation time, and shear modulus of the resulting gel. The hydrogel blends were printed on a custom-made microvalve-based drop-on-demand printing system to evaluate their 3D printability. Furthermore, the stiffness and relaxation of the hydrogels were measured using compression and indentation tests. In cell culture experiments, a tri-culture model was established: for pre-vascularization, the endothelial cells and nasal fibroblasts were integrated into the hydrogels, whereas the surface of the hydrogel was seeded with respiratory epithelial cells.

The gelation properties of collagen could be influenced by consequent pH value control. Furthermore, the resulting agarose-collagen blend exhibited significantly improved relaxation as a prerequisite for cell spreading compared to pure agarose. Nasal fibroblasts were established as supportive cells for angiogenesis and the promotion of respiratory cell differentiation. Besides the fibrin control, it was shown that the composite hydrogel was also able to promote the formation of tubular branched networks. Additionally, the hydrogel surface of fibrin and agarose-collagen provided a suitable growth area for the epithelium proliferation and differentiation after cultivation. In the long-term cultivation, the hydrogel demonstrated mechanical strength and shape stability, which is a decisive precondition for 3D printing of trachea constructs.

In conclusion, a promising novel hydrogel blend enabling angiogenesis as well as the formation of ciliated epithelium with an intrinsic 3D printability was identified and could, for example, serve in the manufacture of *in vitro* 3D trachea models to capture more complex features of disease and drug discovery.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P006

New polymers for additive manufacturing

*C. Hege[1], P. Poh[2], L. Merlat[1], D. W. Hutmacher[3], B. Fischer[1]
[1]ISL, , Saint-Louis, Frankreich
[2]TechnischeUniversität München (TUM), Klinikum rechts der Isar , München, Deutschland
[3]Queensland University of Technology (QUT), Institute of Health and Biomedical Innovation (IHBI) , Brisbane, Australien

For tissue engineering one needs scaffolds that define a structure to proliferate and regenerate the tissue. One of the best methods to produce them is 3D-printing, which allows to fabricate reproducible scaffolds. This has already led to some success, but is still hampered by the shortage of raw material. Only few substances like poly(caprolactone) and natural biopolymers like collagen are used nowadays. This confines the range of achievable mechanical properties and degradation times. The analyzed scaffolds for breast regeneration are often three magnitudes too stiff.

A possible alternative could be copolymers. We examined copolyesters like poly(DL-lactide-co -ethylene brassylate) and poly(DL-lactide-hexadecenlactone). Biodegradable polyesters are normally synthesized with $Sn(octanaote)_2$, but it is in discussion to be cytotoxic.[1] So we analyze enzymes or biocompatible metal salts as alternatives. Iron(III)perchlorate is a good catalyst for ring-opening polymerization of ε-caprolactone[2]. So we also use it for the copolymers, but also other catalysts like enzymes.

With iron(III)perchlorate very flexible poly(DL-lactide-co-ethyene brassylate)polymer could be synthesized. This polymer will be printed and cytotoxicity tests will be made.

References

[1]M. C. Tanzi, P. Verderio, M. G. Lampugnani, M. Resnati, E. Dejana, and E. Sturani, *J. Mater. Sci. Mater. Med.*, 1994, **5**, 393–396.
[2] C. S. Hege and S. M. Schiller, *Green Chem.*, 2014, **16**, 1410–1416.

Figure 1

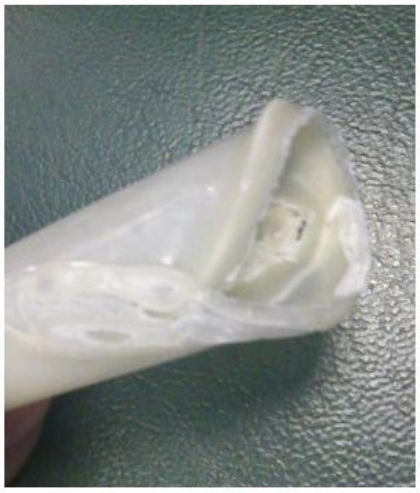


Photo of the poly(DL-Lactide-co ethylene brassylate) polymer, which was catalyzed with iron(III)perchlorate

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P009

Development of Alginate Based Bioink Combined with Re-Engineered Porcine Pericardium for 3D-Printing

M. Schmidt[1,2], *T. Igelspacher[1], A. Rzany[1], R. Detsch[2], A. R. Boccaccini[2], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland
[2]Lehrstuhl für Biomaterialien, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Introduction

3D-printing is a novel approach which combines the 3D-cultivation of cells in bioink and exploits additive manufacturing (AM) techniques to develop 3D-tissue structures. In this context, the bioink acts as a semipermeable membrane and should offer mechanical and biological properties to mimic natural tissues in function, composition and (3D) morphology.

Objectives

Bovine or porcine pericardium has a high content of collagen and additionally contains elastin, glycosaminoglycans and proteoglycans [1,2] and can be used as a component of bioink for 3D-printing approaches in combination with well established alginate based bioink [3].

Materials & Methods

Porcine pericardium is treated in a cryo-cooled homogenizer by strongly accelerating tubes which contain small ZrO_2 beads and a fluid, e.g. 0.9% NaCl solution. The tissue is fragmented by the impact of the spheres. The resulting tissue suspension (PER) is filtered to obtain a defined fragment size range. Subsequently alginate-dialdehyde (ADA) was mixed with PER and finally gelled by calcium chloride. For 3D-printing a three axis bioplotter was used. Beside material characterization, the printing process was evaluated.

Results

By adjusting pressure and printing velocity, different geometries were manufactured. Microstructure was analyzed by scanning electron microscopy and the structural influence of the PER is discussed. Additionally, it could be demonstrated that support structures composed of Pluronic® F127 can be successfully integrated in the print. Further in vitro test will be performed to test the biological functionality of this novel hydrogel matrix.

Conclusion

ADA-PER is a promising candidate for a novel bioink that contains a re-engineered biological material as reinforcement for novel 3D-printing applications.

References

[1] E. Rémi et al. In: Biomaterials Science and Engineering: In Tech (2011), S. 437–455.
[2] A. S. Braga-Vilela et al. J Membrane Biol (2008) 221:15–25
[3] Zehnder, T., Sarker, B., Boccaccini, A. R. & Detsch, R. Evaluation of an alginate–gelatine crosslinked hydrogel for bioplotting. Biofabrication 7, 1–12 (2015).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P010

Optimization of hyaluronic acid/poly(glycidol) hybrid hydrogels for the generation of double-printed cartilage constructs

*T. Böck[1,2], S. Stichler[1], N. Paxton[1], S. Bertlein[1], R. Levato[3], V. Schill[1], J. Malda[3,4], J. Teßmar[1], T. Blunk[2], J. Groll[1]
[1]Universitätsklinikum Würzburg, Abteilung für Funktionswerkstoffe der Medizin und der Zahnheilkunde (FMZ) , Würzburg, Deutschland
[2]Universitätsklinikum Würzburg, Klinik und Poliklinik für Unfall-, Hand-, Plastische und Wiederherstellungschirurgie , Würzburg, Deutschland
[3]University Medical Center Utrecht, Department of Orthopaedics , Utrecht, Niederlande
[4]Faculty of Veterinary Medicine, Utrecht University, Department of Equine Sciences , Utrecht, Niederlande

The use of hydrogels as bioinks for 3D bioprinting makes high demands in terms of printability and cell compatibility of the applied materials. Therefore, the fine-tuning of the physical material properties during bioink development has to be integrated with the specific requirements that ensure the biological function of encapsulated cells, especially with regards to viability and differentiation capacity. To address these challenges, we optimized a poly(glycidol) (PG)-based hydrogel system that was previously shown suitable as bioink for 3D printing [1]. This bioink was combined with allyl-functionalized PG (P(AGE-*co*-G)) as a cell-friendly cross-linker and thiolated hyaluronic acid (HA-SH) to provide adequate biological cues for the generation of 3D-printed cartilage constructs. The cross-linking of the hydrogels with 10 wt.% overall polymer concentration (5 wt.% of each polymer) was achieved by UV-mediated thiol-ene-click reaction between the thiol and allyl groups. In order to adjust the rheological characteristics for extrusion based bioprinting, high molecular weight HA (1.36 MDa; HMW-HA) was added to the hydrogel precursor solution. While the incorporation of HMW-HA led to a lower Young"s modulus and a higher swelling ratio of the hydrogels during the first 24 h, the equilibrium swelling was comparable between all gels after 24 h. To demonstrate the feasibility of the bioink for cartilage regeneration, the chondrogenic differentiation of human and equine mesenchymal stem cells (MSCs) was investigated. Upon incorporation within the gels and chondrogenic differentiation for 21 days in vitro, both cell types exhibited robust chondrogenesis as indicated by staining against cartilage-specific extracellular matrix molecules (e.g. aggrecan and collagen type II) and quantification of glycosaminoglycan and collagen content. In contrast, the addition of HMW-HA did not seem to affect the viability of human MSCs (hMSCs) but impaired chondrogenic differentiation. Further, hMSCs showed good cell viability after the bioprinting process. Finally, the feasibility of the developed bioink for the generation of cartilage constructs was demonstrated by double-printing with poly-ε-caprolactone (PCL), resulting in constructs with enhanced mechanical resistance that closely resembled the mechanical properties of native articular cartilage.

References

[1] Stichler, S. et al. Thiol-ene Clickable Poly(glycidol) Hydrogels for Biofabrication. Ann. Biomed. Eng. **45**, 273-285 (2017)

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P011

Improved alginate based hydrogel systems as model for cancer research studies

*V. Bednarzig[1], A. R. Boccaccini[1], *R. Detsch[1]
[1]Friedrich-Alexander Universität Erlangen-Nürnberg, Lehrstuhl für Biomaterialien , Erlangen, Deutschland

Introduction

Colorectal cancer is the third leading reason for cancer death in the US [1]. The main issue is that this kind of cancer shows only symptoms in advanced stages [2]. Due to that it is important to understand its tumorigenesis 3D cancer models can provide solutions for development of new cancer therapies.

Objectives

The aim of this research was to improve an established alginate-based hydrogel system, to use them in for cancer research. For this propose biofabricated models were designed via 3D bioplotting, focused on the adjustment of the hydrogels" stiffness via different concentrations and gelation ions.

Materials & Methods

As bioink a combination of alginate di- aldehyde (ADA) and gelatin (GEL) was used. Both materials were covalently crosslinked and additionally ionically gelled (with 0,1M CaCl2, 0,1M BaCl2 and combinations of it). While ADA the partially oxidized product of alginate is used because of its controllable degradation behavior and higher porosity, gelatin is applied due to its ability to bind cells, through integrin-binding sites, caused by the RGD- sequence of collagen [3]. Therefore in vitro evaluation of the biofabricated constructs HCT116 cells were cultured for 3 weeks.

Results

Measured values of the young"s modulus showed that the stiffness (from 21,5 kPa up to 45 kPa) can be tuned by high ADA content and the use of BaCl2. The design of the hydrogels also affected the cell behavior. Low numbers of dead cells and high cell proliferation rates were analysed in the different hydrogels after 21 days, whereas hydrogels crosslinked with BaCl2 induced superior HCT116 cell behavior compared to the ones gelled with CaCl2.

Conclusion

This study revealed that ADA-GEL crosslinked with BaCl2 enhanced the stiffness of the hydrogel and induced higher the adhesion, proliferation and viability of the HCT116 cancer cells compared to the used of CaCl2. Through the combination of varying gel contents and different gelling ions it is possible to adjust the required young"s modulus to mimic natural soft tissues.

References

[1] Siegel R, Desantis C, Jemal A (2014), CA Cancer J Clin 64(2): 104-117
[2] Ivanovska J, Zehnder T, Lennert P et al. (2016), Tissue Eng Part C Methods 22(7): 708-715
[3] Sarker B, Papageorgiou DG, Silva R et al. (2014), J. Mater. Chem. B 2(11): 1470

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P012

Fabrication of drug-loaded calcium phosphate cement scaffolds by core/shell 3D printing.

*A. R. Akkienni[1], M. Brückner[1], T. Ahlfeld[1], A. Lode[1], M. Gelinsky[1]
[1]Universitätsklinikum Dresden, Zentrum für Translationale Knochen-, Gelenk- und Weichgewebeforschung , Dresden, Deutschland

Additive manufacturing (AM) methods for tissue engineering (TE) provide excellent tools for fabrication of custom-designed 3D structures. Among the many AM methods, 3D extrusion printing allows the fabrication of constructs, having clinically relevant sizes, also made of more than one biomaterial by using multi-channel printing. Consequently, complex patterns of drug/cell delivery or cell reorganization can potentially be mimicked in such 3D constructs *in vitro*. Thus after implantation of these constructs, the integration and repair at the defect site *in vivo* can be more efficient.

Based on multi-channel 3D extrusion printing, a method to integrate two different naturally derived biopolymers that are spatially separated in a single strand – arranged as core and shell (C/S) (Fig: 1A) – has been recently described [1]. In this work, low concentrated biopolymers (as core) and pasty calcium phosphate cement (CPC; as shell) have been combined to fabricate C/S scaffolds intended for bone TE applications. Hardening of the scaffolds was achieved by two different processes i.e. set in humidity and in water [2]. These scaffolds exhibited novel properties as an effect of the intrinsic properties of the individual materials. Higher porosity was observed in C/S scaffolds compared to conventional CPC scaffolds. Due to the contact of the CPC with the hydrogel-core, the cement setting started already during extrusion that led to improved shape fidelity of the scaffolds. Though not significant, Young"s modulus was lower for C/S scaffolds. Low concentrated biomaterials used in the core and mild processing conditions enabled encapsulation of sensitive components such as growth factors. As a model protein Bovine serum albumin (BSA) and Lysozyme were loaded in the core materials (3 % Alginate and 1.5 % Chitosan) and the protein release was probed based on the setting conditions. A sustained release of vascular endothelial growth factor A (VEGF-A) was observed and the bioactivity was preserved (confirmed by proliferation assay and *in vitro* angiogenesis assay) when loaded in the core. Furthermore, C/S bioprinting was probed to fabricate perfusable hollow strand 3D scaffolds to colonise the inner lumen with different cell types intended to be used for vascular TE applications.

References:

[1] Akkineni, A. R *et al.* (2016) *Biofabrication 8*, 45001.
[2] Akkineni, A. R *et al.* (2015) *Acta Biomater.* 27, 264-274

Figure 1

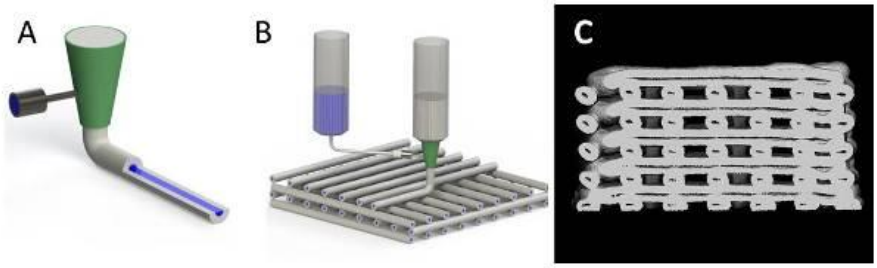


Figure 1: Schematic diagram of Coaxial needle and C/S scaffold printing (A & B). µCT image of a CPC C/S scaffold (C).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P013

Fabrication of Sinusoidal-like Scaffolds for Tendon Tissue Engineering Applications

*A. Youssef[1], A. Hrynevich[1], H. Yin[2], G. Hochleitner[1], J. Groll[1], D. Docheva[2], P. D. Dalton[1]
[1]Univeristy Hospital Würzburg, Functional Materials in Medicine and Dentistry , Würzburg, Deutschland
[2]University Hospital Regensburg, Experimental Trauma Surgery , Regensburg, Deutschland

Introduction: Tendon injuries are a source of significant morbidity to patients. Melt electrospinning writing (MEW) is used to produce defined fiber-based scaffolds where shape, fiber diameter and layer-by-layer stacking can be controlled to micron-scale printing resolutions [1]. Native tendon, which has a collagen extracellular matrix, has a highly organized "crimped" ultrastructure, and this is reflected in the mechanical properties of this tissue. Here we describe the direct-writing of "sinusoidal" scaffolds for tendon tissue engineering. Tendon cells are mechanoresponsive [2] and we used a subtype of them, tendon stem/progenitor cells (TSPCs), which are important for tendon development and repair [3].

Materials and methods: A custom-made MEW device was used to print medical-grade polycaprolactone melt. The G-code for the collector movement allowed an oscillatory moving pattern. Straight fiber scaffolds were also produced as controls. Characterization of the scaffolds was performed by scanning electron microscopy and mechanical testing. TSPCs were seeded with a density of 3x105 cells per scaffold inside a 12-well cell culture plate. Live/dead staining was performed at days 1, 3 and 7.

Results: The sinusoidal fibers had a diameter of 15 µm and a crimp angle of 25-35°. Straight scaffolds had the same fiber diameter. The stress/strain curve showed a toe region for sinusoidal scaffolds compared to straight ones [Fig. 1]. Live/dead staining showed attachment of TSPCs on day 1 and proliferation till day 7. The cells were generally aligned with the overall direction of the fibers in both scaffold types [Fig. 2].

Conclusion: It was possible to print sinusoidal MEW scaffolds with fiber diameter approximately equivalent to the primary collagen bundle in a tendon. The "toe region" in the stress/strain curve of sinusoidal scaffolds is of similar shape to that of native tendon while TSPCs could attach and proliferate and align along the direction of the fibers. Further exploration of the effects of crimped fibers on TSPCs is planned by checking gene expression of tenogenic markers.

Fig. captions:

Fig. 1: SEM image and stress/strain curve of sinusoidal (A&B) and straight (C&D) scaffolds.

Fig. 2: Live/Dead staining after 7 days in (A) sinusoidal and (B) straight scaffolds.

References

[1] Brown TD, et al. Prog Polym Sci 2016 56: 116-166.
[2] Walden G, et al. Tissue Eng Part B Rev 2017 23(1): 44-58
[3] Docheva D, et al. Adv Drug Deliv Rev 2015 84: 222-239

Figure 1

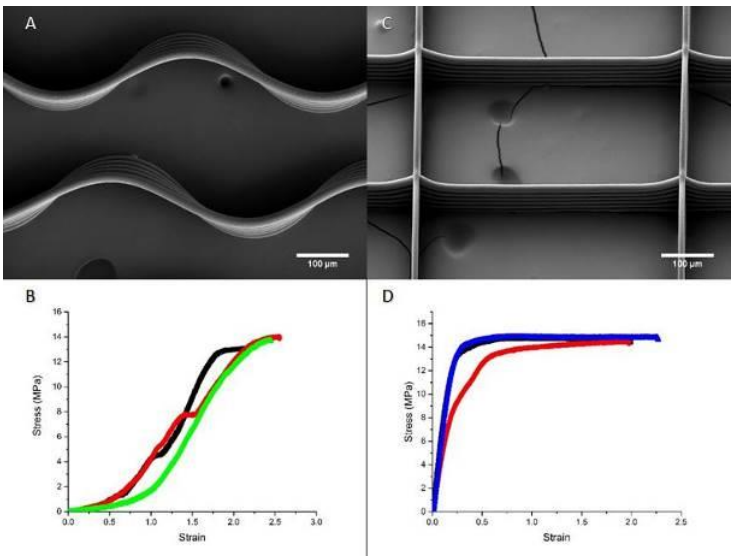
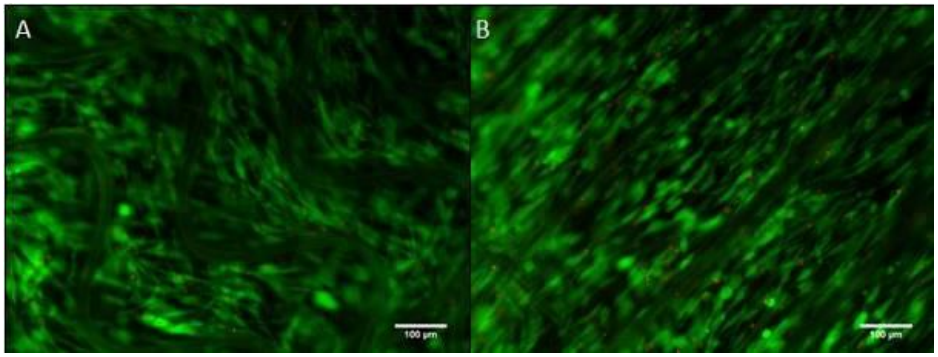


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P014

Biomimetic *in vitro* analysis of the fatigue behaviour of cyclically loaded UHMWPE structures for orthopedic implantation

*D. Porchetta[1], R. Scholz[1], M. Knyazeva[1], F. Walther[1]
[1]TU Dortmund, WPT , Dortmund, Deutschland

Introduction

An important goal of research in the field of biomaterials lies in the progressive substitution of in vivo tests with suitable *in vitro* experiments. This will lead to the avoidance of the ethical issues bound to the sacrifice of animals for the sake of scientific research. Another advantage of transferring the experiments to the *in vitro* field is the possibility of accurately control the boundary conditions and the experimental parameters in order to reduce, or eliminate, the need of validation tests involving animals. With the aim to reduce the amount of needed *in vivo* studies for the determination of the long-term durability of ultra high molecular weight polyethylene (UHMWPE) under fatigue loading in a biological environment, a short-time *in vitro* test procedure using instrumented fatigue load increase tests with superimposed environmental loading has been developed at WPT.

Methods

Within the framework of developing a suitable bio simulative testing rig, several samples of UHMWPE were investigated under static and cyclical loading related to the foreseen implantation site. The mechanical tests are meant to adhere to the biomimesis principle of testing by taking place in a simulated body fluid (SBF) environment. FT-IR spectroscopy is implemented in order to characterize the degree of damage of the UHMWPE samples as a function of the number of cycles.

Results

Due to the developed test stand, test runs simulating cyclically loaded implantation sites are possible and thus more precise predictions of the material behaviour (aging, oxidation, fatigue) are rendered possible. The characterisation of the sample damage by use of FT-IR spectroscopy results helpful but not decisive in the correlation with the fatigue lifespan. None of the implemented SBF showed a higher degree of influence in the degradation behaviour of the analysed samples. Over the entire period, the measured pH values were always within the physiologically tolerable range.

Conclusions

By developing a biosimulative test rig for the fatigue life analysis of UHMWPE orthopedic implants it is possible to improve the closeness as well as the predictiveness, to a certain extent, of a material behaviour in an in vivo environment. By means of further testing and development, a reproducible and ethically sustainable alternative to animal experiments could be achieved, allowing for a higher degree of pre-implantation behaviour prediction.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P016

The photoelectrochemical response of coated and uncoated steel acupuncture needles during UV exposure

*T. Noack[1,2], C. Schille[2], E. Schweizer[2], H. Mantz[1], J. Geis-Gerstorfer[2]
[1]Hochschule Ulm, , Ulm, Deutschland
[2]Universitätsklinikum Tübingen, Medizinische Werkstoffkunde und Technologie , Tübingen, Deutschland

Acupuncture is an essential feature of the traditional Chinese medicine (TCM), one area of the alternative medicine, in which thin needles are inserted into special body points to alleviate mainly chronic pains. Mostly thin needles made by pure steel are used. Little is known about the photocatalytic activation of TiO_2 coated steel needles, which is intensively studied as a material for antimicrobial and self cleaning applications under UV exposure [1].

In this work the photoelectrochemical activation of different acupuncture needles in isotonic saline solution was investigated. Pure steel acupuncture needles (n=5) and needles additionally coated with TiO_2 semiconductor alloy in Anatas modification (n=5) - with and without UV exposure were compared (Hönle; 382nm; UV exposure: 1 min; power: 44mW/cm²; distance to sample: 187mm). A UV-transparent electrochemical cell was used (PAR 263A; Software: M352). It consists by a saturated calomel electrode as reference electrode, graphite as counter electrode and the needle as working electrode. Open-circuit-potential measurements over 50 minutes and potentiostatic measurements at 0mV~Ecorr over 10 minutes were performed. After half of time the UV light was turned on for 1 minute.

UV exposure has a strong effect on the electrical potential of TiO_2 on stainless steel needles compared to pure stainless steel needles- peak heights: 329mV vs. 41mV (Figure1). UV exposure has no effect on the rest potentials of uncoated needles before and after exposure. However the rest potentials of coated samples with TiO_2 differ considerably - Ecorr before exposure 78.4mV and afterwards 37.6mV. The photocurrent with TiO_2 coated samples peaks to 21.6µA with UV irradiation, but exhibits no effect on stainless steel needles (Figure 1). To detect any corrosion acupuncture needles were additionally imaged by a scanning electron microscope (SEM). No remarkable visible defects on the surface could be observed (Figure 2).

Electrochemical measurements with different acupuncture needles were performed and it was shown that the photoelectric effect of acupuncture needles can be improved due to coating with TiO_2. Our findings could be the basic of further research for the influence of materials on medical effects of acupuncture.

References

[1] *S. Daviðsdóttir et. al, "Investigation of photocatalytic activity of titanium dioxide deposited on metallic substrates by DC magnetron sputtering," Surface and Coatings Technology, vol. 216, pp. 35–45, 2013.*

Figure 1

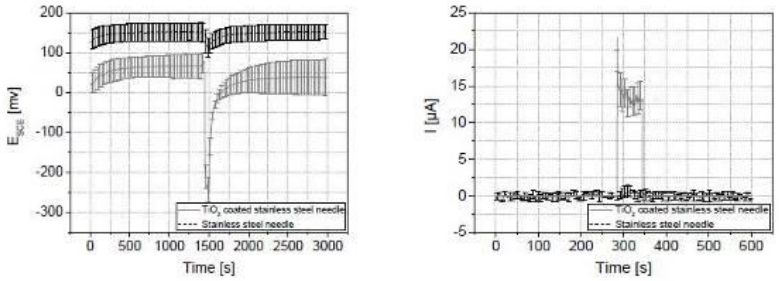


Abb.1: Open-circuit-potential and potentiostatic measurements of coated and uncoated needles during UV exposure for 1 min

Figure 2

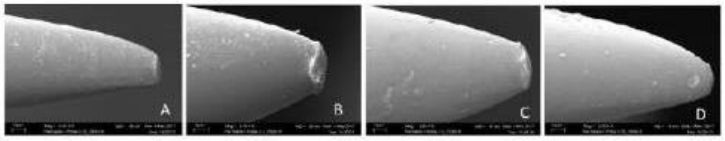


Abb.2: SEM images of the needle tips before and after UV exposure - uncoated (before: A, after: B) and coated (before: C, after: D), 5000x magnification

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P017

The particle-induced cell migration assay (PICMA): An in vitro assay to assess the effect of inflammatory particle

*A. Rostek[1], I. Föhring[2], K. Loza[1], A. Brik[2], N. Rosenkranz[2], C. Sengstock[3], M. Köller[3], T. Brüning[2], G. Johnen[2], G. A. Westphal[2], J. Bünger[2], M. Epple[1]
[1]University Duisburg-Essen, Inorganic Chemistry and Center for Nanointegration Duisburg-Essen (CeNIDE) , Essen, Deutschland
[2]Institute for Prevention and Occupational Medicine of the German Social Accident Insurance, Institute of the Ruhr-University Bochum (IPA), , Bochum, Deutschland
[3]Bergmannsheil University Hospital/Surgical Research, Ruhr-University Bochum, , Bochum, Deutschland

Introduction

The interaction of micro- and nanoparticles with cells and tissue is an important topic in current biomedical research, e.g. in the case of aseptic loosening of endoprostheses. The underlying pathophysiologic mechanism of particle toxicity is inflammation. We have established a new *in vitro* model to investigate chemotaxis together with the assessment of inflammatory mediators in response to the exposure of micro- and nanoparticles. The chemical composition as well as other particle properties (e.g. shape, size, and charge) strongly influence the biological response in a complicated way. To elucidate these particle effects apart from the chemical composition, we have prepared $BaSO_4$ in the form of nano-, submicro- and microparticles as bioinert control and studied the cellular response.

Methods

NR8383 rat macrophages were incubated with selected and well-characterized particles (quartz, silica, rutile, anatase, carbon black, and $BaSO_4$). The supernatants of the incubated cells were used to study the migration effects on dHL-60 cells in the PICMA. In addition, the loss of cell adherence concerning particle exposure was associated with a decreasing cell index determined by real-time cell analysis as indicator for the cytotoxicity. The chemotaxis assay, based on the Boyden chamber technique, was carried out in parallel to transcription analysis of inflammatory mediators in response to the particle exposure.

Results

$BaSO_4$ and TiO_2 (anatase) particles showed only a weak impact on the cell migration and the cell adherence. In contrast, nano-silica ($SiO_2 \cdot n\ H_2O$) and nano-rutile (TiO_2) particles acting strongly showing an increase in cell migration and led to a high cytotoxicity. The transcription of pro-inflammatory mediators (e.g. CXCL8, TNF-α) was most pronounced by nano-silica and quartz (SiO_2) particles. $BaSO_4$ particles (nano, submicro and micro) did not induce adverse effects, making them suitable as inert reference material to study cell-particle interactions.

Conclusion

The presented PICMA uses permanent cell lines, is highly reproducible and is clearly able to distinguish between particles with different inflammatory potential. The assay enables the investigation of cell migration and measures in parallel the transcription of pro-inflammatory mediators. Moreover, it is also useful to study particle-induced inflammatory signaling and the influence of different particle parameters (like size and shape) as a standardized inflammation assay.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P018

Rezellularisierungsstrategien für eine zellbefreite menschliche Tractus iliotibialis Matrix mit allogenen Tractus Fibroblasten und mesenchymalen Stromazellen

*C. L. Gögele[1], S. Schwarz[1], B. Ondruschka[2], N. Hammer[3], G. Schulze-Tanzil[1]
[1]Paracelsus Medizinische Privatuniversität, Anatomie , Nürnberg, Deutschland
[2]Universität, Rechtsmedizin , Leibzig, Deutschland
[3]University Otago School of Medical Sciences , Anatomie , Otago, Neuseeland

Ziel

Das Ziel der vorliegenden Studie war es, eine erfolgreiche Rezellularisierung eines dezellularisierten Tractus iliotibialis Gewebes mit allogenen Tractus iliotibialis Fibroblasten und mesenchymalen Stromazellen (MSCs) zu erreichen und eine Besiedlung mit dreidimensionalen Sphäroiden zu etablieren.

Methoden

Die Probenentnahme fand immer im Zuge einer Obduktion statt und insgesamt wurden die Gewebestücke von sechs Patienten entnommen. Die Dezellularisierung erfolgte mit einem an das Tractus-Gewebe angepaßten Protokoll mit u.a. Einfrier-/Auftauzyklen und einem Detergenz-basierten Dezellularisierungspuffer. Die Zellbefreiung wurde mit histologischen Färbungen (Hämatoxylin-Eosin, Alcianblau und DAPI) überprüft. Die Zytokompatibilität der ECM wurde vor der Wiederbesiedlung über einen Zytotoxizitätsstest mit ECM Extrakten mit der murinen Fibroblastenzelllinie L929 überprüft. Für die Rezellularisierung mit Tractus Fibroblasten und MSCs wurden drei verschiedene Strategien eingesetzt: eine statische Kultur sowie eine dynamische Suspensionskultur. Zusätzlich wurde eine Besiedlung mit präformierten dreidimensionalen Zellsphäroiden bei sich anschließender dynamischer Kultivierung etabliert. Zelladhärenz, -überleben und -verteilung in der rekolonisierten ECM wurde mit mikroskopischen Untersuchungen (Lebend-Totfärbung und histologischen Färbungen) überprüft.

Ergebnisse

Die Dezellularisierung war erfolgreich: es konnten keine verbliebenen Zellen mehr in der Tractus ECM nachgewiesen werden. Die Testung von ECM-Extrakten in L929 Fibroblasten ergab keine Hinweise auf Zytotoxizität durch etwaige Rückstände des Dezellularisierungspuffers. Mit allen Rezellularisierungsstrategien konnte eine Kolonisierung der ECM mit Tractus Fibroblasten und MSCs erreicht werden, wobei die dynamische Kultur reproduzierbar am erfolgreichsten war. Die Zelladhärenz bei Besiedlung mit MSCs war im Vergleich zu der Verwendung von Tractus Fibroblasten geringer. Auch die Sphäroid-basierte Technik kombiniert mit der dynamischen Kultur war mit Tractus Fibroblasten erfolgreich – sie ermöglicht zukünftig eine sehr gezielte lokale Besiedlung mit Zellen, die vordifferenziert werden können und ihre eigene frisch produzierte ECM mit in das Konstrukt einbringen.

Schlussfolgerung

In der Studie konnte ein vielversprechender Ansatz für die Rekolonisierung einer zellbefreiten Tractus iliotibialis ECM mit allogenen MSCs und Tractus Fibroblasten entwickelt werden.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P019

Adsorption of Proteins on Surfaces

*B. Arlt[1]
[1]Anton Paar Germany GmbH, Nanostrukturanalyse , Ostfildern, Deutschland

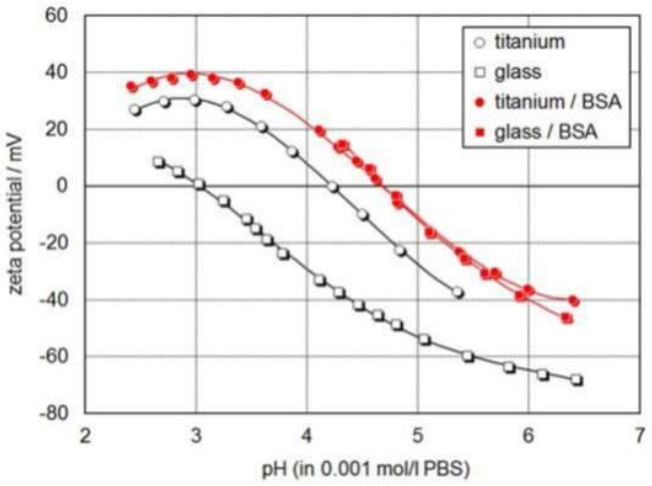
The acceptance of implant materials made of stainless steel or titanium by the human body depends on the biocompatibility of the outermost material surface. Various surface properties determine its biocompatibility. Surface charge drives the electrostatic forces that attract proteins as a prerequisite for the osseo-integration, e. g., of tooth implants. On the other hand, desired electrostatic repulsion prevents adhesion of proteinaceous moieties that initiate infection.

Knowledge of the surface is therefore of paramount importance for the development and qualification of biomaterials for implantation. The zeta potential gives information about the surface charge at physiological pH and the chemistry of surface functional groups.

The sign and magnitude of the zeta potential let us estimate the electrostatic interaction between the solid surface and a charged species dissolved in the surrounding aqueous solution. Being a surface-sensitive parameter, the zeta potential is best suitable to indicate adsorption and desorption processes. Changes in the surface properties upon adsorption are indicated even if the driving force for adsorption is not of electrostatic nature.

The zeta potential analysis combines the measurement of adsorption kinetics with the validation of the chemistry of the adsorbed surface layer. In this presentation we will shed some light on the adsorption of albumin (BSA) on a glass and titanium surface. By help of electrophoresis measurements it is proven, that both surfaces are entirely covered by BSA: The zeta potential value of the surfaces after the adsorption process shows the same values which counts for the pure BSA in a buffered solution.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P020

Development of Protein-Resistant Surface Functionalizations for Biomaterials

*J. Lehnfeld[1], J. Blahnik[1], R. Müller[1]
[1]Universität Regensburg, , Regensburg, Deutschland

Adsorption of proteins on surfaces crucially influences the performance of biomaterials in the human body [1, 2]. Processes, such as thrombosis and inflammation are governed by the initially formed protein layer [2]. Understanding the influence of surface properties on protein adsorption as well as the development of protein-resistant surfaces is thus vital in order to improve the functioning of biomaterials.

For this purpose, a variety of novel surface modifications was developed. Silicon or silica based model substrates were functionalized with self-assembled monolayers (SAMs) via silane chemistry. These SAMs served as a basis for the subsequent immobilization of various reagents with terminal amine groups. In summary, aliphatic amines and amidoamines with oligomeric, polymeric or dendritic structure were investigated (Figure 1).

Figure 1: Overview of the surface modifications: (a) linear poly(ethylene imine) (PEI) polymer, (b) poly(propylene imine) (PPI) dendrimers of generation 2 or 4, (c) N,N'-bis(3-aminopropyl)-1,3-propanediamine (APD), (d) linear polyamidoamine (PAMAM) polymer or (e) PAMAM oligomer.

Apart from physicochemical properties, protein adsorption on those surfaces was investigated with the bicinchoninic acid (BCA) assay. Adsorption isotherms were recorded with varying concentrations (1-50 mg mL^{-1}) of human serum albumin (HSA, pI 4.7) and lysozyme (pI 10.9). PPI dendrimer or PAMAM oligomer coated surfaces showed poor protein-resistance with respect to at least one of the proteins, whereas APD, PEI or PAMAM polymer covered surfaces exhibited protein-repellent properties. Furthermore, protein adsorption from undiluted physiological fluids, human whole saliva and foetal bovine serum (FBS), was examined. In these experiments, non-fouling properties could again be observed for APD, PEI or PAMAM polymer modified substrates (adsorption < 20 ng cm^{-2} from saliva and < 80 ng cm^{-2} from FBS). Comparing all protein adsorption tests, PEI polymer functionalized surfaces exhibited the best protein-repellent properties and are thus promising surface coatings for further application.

Figure 2: Protein adsorption onto coated silica beads from protein solutions (50 mg mL^{-1}, left) or physiological fluids (right). Medians with 25% and 75% quartiles as error bars are provided.

References

[1] J. M. Courtney, N. M. K. Lamba, S. Sundaram, C. D. Forbes, *Biomaterials* **1994**, *15*, 737–744.
[2] I. Banerjee, R. C. Pangule, R. S. Kane, *Advanced Materials* **2011**, *23*, 690–718.

Figure 1

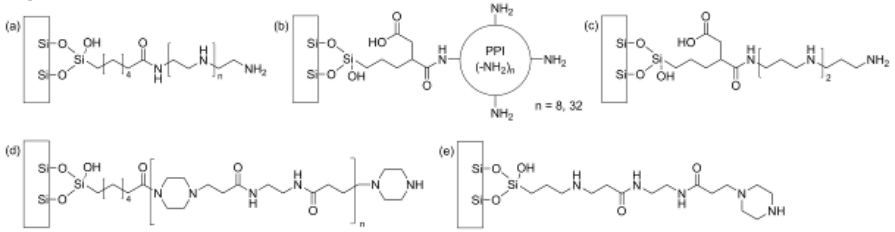
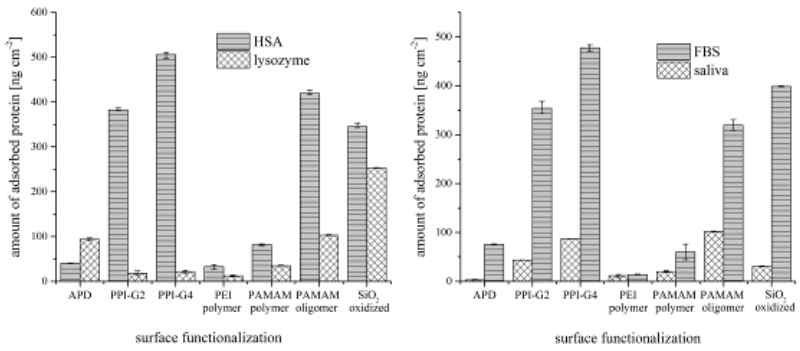


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P021

Morphological and micromechanical analysis of human bone

*J. Klehm[1], S. Henning[1], M. Menzel[1], A. Thormann[1], J. Brandt[2]
[1]Fraunhofer Institute for Microstructure of Materials and Systems IMWS, Biological and macromolecular materials , Halle (Saale), Deutschland
[2]Eisenmoorbad Bad Schmiedeberg GmbH, Orthopädische Rehaklinik 1 , Bad Schmeideberg, Deutschland

Introduction

Bone is a hierarchically structured material. The comparison of fracture and crazing behavior of healthy and diseased bone is of special interest, raising the question in which way pathological changes modify especially the micro- and nanostructure of bone and influence micromechanical processes and increase the risk of fracture. This can lead to a better understanding of the effects of aging, diseases and drugs on tissues, and may help developing a scientific basis for tissue engineering.

Methods

Human femoral heads that had to be removed during total hip replacement surgery after trauma or due to systemic diseases like osteoporosis, osteoarthritis or osteonecrosis were processed according to standard histological procedures to analyze the morphology. For the description of micromechanical processes, different approaches for the preparation of bone samples have been selected. Light microscopic investigations were performed to identify the area of investigation and to detect microcracks. Low Vacuum scanning electron microscopy (Low-Vac SEM) and transmission electron microscopy (TEM) was applied for the micro- and nanoscopic characterization of microcracks and craze-like deformation zones.

Results

Light microscopy and SEM investigations of healthy, osteoporotic and ostenecrotic bone shows different microstructural characteristics. The measurements of HA crystals of TEM images have shown independent of age and clinical findings constant values. The typical nanostructure of healthy bone is characterized by mineralized collagen fibrils consisting of plate-like HA crystals embedded in a collagen matrix. In necrotic bone areas with a disordered nanostructure (fig. 1) could be found, which may lead to a deterioration of mechanical properties and increased risk of fracture. The SEM and TEM images show craze-like structures at the crack tip. It reveals that the crack is bridged by mineralized collagen fibrils. Based on this investigations, initial stages of microcrack formation and growth are associated with an intensive fibrillation and nano-cavitation (fig. 2).

Conclusion

Independent of the preparation pathways, the occurrence of craze-like deformation zones in bone is observed. The edges of microcracks are bridged by mineralized collagen fibrils of a typical diameter of 100 nm. It can be assumed that crazing is a universal phenomenon that takes place at initial stages of microcrack formation and growth.

Figure 1

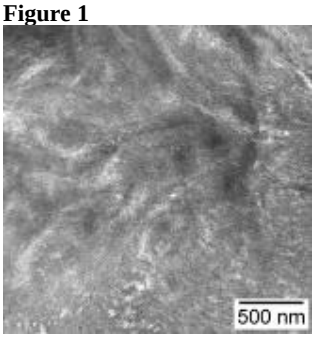
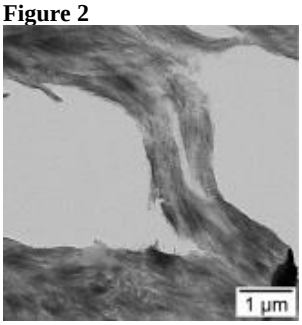


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P022

Vicinal, double chemoselektive biofunctionalization of polyoxazolines via NCL and thiol-ene

*F. Pinzner[1], J. Groll[1]
[1]Universitätsklinikum, FMZ , Würzburg, Deutschland

We present a way of chemoselective and vicinal coupling of hydrophilic polymers and biomacromolecules via *native chemical ligation* (NCL) and subsequent thiol-ene reaction.

The linkeage of three different components at one reaction site is desirable in terms of chemoselectivity, synergistic effects and cooperativity. Using polymers to *vicinally and chemoselectively* couple them with biomacromolecules such as sugars, proteins or peptides is quite interesting, since it opens a wide field of conceivable applications, such as fluorencence labelling or substrate recognition.

As a proof of principle, the sequence of reactions is displayed on a low molecular level. Starting from the literature-orientated synthesis of a thioester[1], we couple the product to a cysteine-functionalized reactant via NCL. Besides from being a reliable and commonly used coupling method,[2] one of the advantages of the NCL is the formation of a free thiol group in the reaction product. This allows us to use them for an ensuing *thiol-ene click reaction* with allyl-functionalized substrates. Since the resulting free thiols from the NCL reaction tend to form dimers through oxidation, a way of reducing those disulfides is also presented.

The subsequent step is to transfer this set of reactions to a macromolecular level. As the main component, we use poly(2-oxazoline) (POx) polymers, that we synthesize bearing either one or multiple thioester-, cysteine- or allyl-functionalities. POx polymers are potential alternatives to the well established poly(ethylene glycol) (PEG) for biomedical applications.[3] They possess similar biocompatibility, but can, in contrast to PEG, be functionalized along the polymer chain.[4] This provides us a set of functionally adjustable polymers that can be used in further reactions, such as coupling with peptides and/or sugars.

The final aim is to be able to connect three different components (polymer + peptide + sugar) chemoselectively at the very same binding site and use the system for substrate recognition with enzymes such as Galectin-1.

References

[1] L. Markey, S. Giordani, E. M. Scanlan, *The Journal of Organic Chemistry* **2013**, *78*, 4270ff.
[2] C. P. R. Hackenberger, D. Schwarzer, *Angewandte Chemie* **2008**, *120*, 10182ff.
[3] R. Luxenhofer, Y. Han, A. Schulz, J. Tong, Z. He, A. V. Kabanov, R. Jordan, *Macromol Rapid Commun* **2012**, *33*, 1613ff.
[4] M. Schmitz, M. Kuhlmann, O. Reimann, C. P. R. Hackenberger, J. Groll, *Biomacromolecules* **2015**, *16*,1088ff.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P023

The hen's egg test on the chick area vasculosa (HET-CAV) as a versatile alternative test model to determine the biocompatibility of biomaterials

*P. Warncke[1], Y. Poetzinger[1], M. Rabel[1], R. Müller[2], O. Stranik[2], W. Fritzsche[2], D. Fischer[1]
[1]Friedrich-Schiller-University Jena, Department of Pharmaceutical Technology , Jena, Deutschland
[2]Leibniz Institute of Photonic Technology, Department of Nanobiophotonics , Jena, Deutschland

Question

Chicken-based test systems using eggs are common in medical and toxicological research for many years and some of them are accepted by authorities. Especially in the early stages of the development of new materials or formulations, the irritation potential or toxicity has to be assessed thoroughly. For this purpose, the hen's egg test on the chick area vasculosa (HET-CAV) offers a dynamic planar vascular network system for the assessment of local or systemic effects of materials or particles.

Methods

Fertilized hen's eggs were incubated for 72 h and transferred into petri dishes to obtain the planar chick area vasculosa (CAV). Different biomaterial samples varying by size, shape and surface characteristics were tested in various ways using the HET-CAV. Besides the local application of macroscale samples (bacterial nanocellulose fleeces), also systemic injections of micro- or nanoparticles (acidic hydrolyzed nanocellulose whiskers or biomaterial coated iron oxide particles) were performed to investigate the irritation potential of various materials for up to 24 h. Therefore, hemorrhage, vascular lysis, thrombosis and embryonic lethality were assessed as relevant toxic reactions after the application or injection of materials. Furthermore, the flow profile pattern of particles within the blood stream was observed using fluorescence microscopy and video analysis of the particles after systemic injections.

Results

The HET-CAV was established to distinguish between toxic or biocompatible materials by macro- or microscopic analysis after local or systemic administration. A scoring system was established to evaluate the influence of relevant toxic reactions after the application of various samples. Data obtained from the *ex ovo* HET-CAV correlated with data from static 2D-cell culture toxicity assays (MTT assay) *in vitro*. Besides the toxicological profile, further fluorescent microscopic investigations after systemic particle injections demonstrated the influence of the particle surface characteristics on the particle flow in the blood stream. Due to its dynamic blood flow, the hen's egg vascular network imitates the complex *in vivo* situation and could overcome limitations of static cell culture assays.

Conclusions

In conclusion, the HET-CAV offers a flexible alternative test system to investigate a variety of different materials in a biological surrounding according to the 3R concept of animal testing

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P024

Warm Pressing of Native Porcine Pericardium

*S. Weingärtner[1], A. Rzany[1], B. Hensel[1]
[1] Max-Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Bovine and porcine Pericardium are frequently used as biomaterials for various application in medicine. [1] The thickness and homogeneity of thickness are important for implants like biological heart valves. To this goal the pericardium is pressed at elevated temperatures. We present an analysis of various properties that are influenced by the treatment.

Porcine pericardium is obtained from the slaughterhouse and processed within less than 48h. The pericardium is mechanically prepared to only obtain the pericadium fibrosum and lamina parietalis. Dehydration is prevented by moistening with 0.9 % NaCl.

After preparation the tissue is pressed with a force of 98 kN for 20 minutes with temperatures ranging between 26°C and 80°C and stored in 0.9 % NaCl. Part of the pericardium is stored in 30 % glycerine solution before pressing.

As reference native pericardium is used.

Parameters determined are water content by weight difference before and after vacuum drying, denaturation temperature obtained from Differential-Scanning-Calorimetry, mechanical properties obtained from with uniaxial tensile tests, tensile stress and strain; and tissue structure by Scanning-Electron-Microscopy (SEM).

Pressing of the pericardium does not affect the water content. Denaturation temperature does not change after pressing at temperatures below the denaturation temperature. Thickness decreases and area increases irreversibly through pressing. The mechanical properties are not significantly changed. SEM-images show that for higher temperature during pressing the cavities between the fibres disappear, also the fibre bundles and layers of fibres merges. These effects are reduced if the pericardium is stored in a glycerine solution before pressing.

The results show, that the pressing has a permanent effect on the pericardium. The fact, that water content, denaturation temperature and mechanical properties are not significantly changed is an indication, that the collagen fibres of the tissue are not destroyed. The reduction in thickness and increase in area indicate that the Extracellular Matrix (ECM) is affected and the microscopic structure is remodel by altering the size and distribution of voids.

References

[1] Rémi E. et al; Biomaterials Science and Engineering DOI:10.5772/24949

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P025

Optimized Processing of Crosslinked Porcine Pericardium Patches with Homogenous Thickness

*S. Weingärtner[1], M. Erras[1], A. Rzany[1], B. Hensel[1]
[1] Max-Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

In the last decade, the TAVI method (Transcatheter Aortic Valve Implantation) has been established as durable alternative to surgical aortic valve replacement in patients with high operative risk [1]. The material used to build these prosthesis is commonly porcine pericardium with a native thickness between 0.1 and 0.15 mm. The tissue has to be processed with glutaraldehyde (GA) to improve its mechanical properties and to provide the necessary biological stability in the human body [2]. Usually, the native thickness increases by about 70% during the crosslinking in GA solution. We present a method to decrease the thickness of the porcine biomaterial while its other properties are conserved or even improved.

The basic idea is to apply uniaxial pressure to the tissue while being stored in 0.5% GA solution prepared from Dulbecco´s Phosphate Buffered Saline and Glutaraldehyde, EM Grade, 50%. For this purpose a simple setup is used that consist of two plates that clamp the tissue during exposition to GA. To make sure that a sufficient amount of GA reach the tissue between the plates, a polymer sponge (PUR, density 40 – 60 kg/m3) is inserted between the tissue and the upper plate. Different parameters are examined regarding thickness, homogeneity of thickness, denaturation temperature and mechanical properties.

Using the described method, it is possible to obtain crosslinked tissue with a thickness of 0.08 +-0.02 mm and even less. Homogeneity is improved if compared to the native tissue. The denaturation temperature obtained from Differential Scanning Calorimetry has shown that crosslinking can only be assured by using perforated plates on the side of the sponge, so that the GA can reach the whole tissue.

References

[1]Athappan, G. *et al*; JACC Vol. 61, No. 15, 2013; 1585–95
[2] Rémi, E. *et al; 2011 DOI:10.5772/24949*

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P026

3D-Crosslinking of Porcine Pericardium with Glutaraldehyde Solution

*S. Weingärtner[1], D. Niopek[1], A. Rzany[1], B. Hensel[1]
[1] Max-Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Component parts with a macroscopic 3D-structure can have significantly different properties than the basic material itself. A well-known example for this effect is corrugated cardboard. In this contribution we use porcine pericardium as the base material and fixation with glutaraldehyde in 3D-molds to create the 3D-structure.

Native porcine pericardium is mechanically processed while permanently moistening it with 0.9% NaCl solution in order to prevent irreversible dehydration. A glutaraldehyde solution with DPBS is prepared for crosslinking. Two matching 3D-molds (made from PLA) with the shape of a wave were printed with a 3D-printer and the pericardium was carefully inserted between them. Air bubbles between the mold and the pericardium were avoided. Both parts were loosely fixed to keep their relative positions when floating in the glutaraldehyde, without hindering the glutaraldehyde to reach the whole pericardium. For verification of the crosslinking the denaturation temperature was determined using differential scanning calorimetry (DSC). [2]

It is shown show that it is possible to transfer the 3D-wave structure to the porcine pericardium by the presented method. The denaturation temperature is the same as obtained for freely cross-linked pericardium. The resulting tissues, with or without the 3D-structure, are similar in colour and feeling when touched. The 3D-structure is impressed permanently. When straightening it carefully by pulling it returns to its initial position by itself when let free, just like a spring. The initial shape is regained faster when the tissue is freely floating in liquid. Storing the tissue subsequently in glutaraldehyde solution does not change the 3D-structure.

References

[1] Julien et al; Biomaterials 18 (1997) 605-612
[2] Damink et al.; J. Mater. Sci. Mater. Med. (1995) Vol 6, 8, 460-472

Figure 1

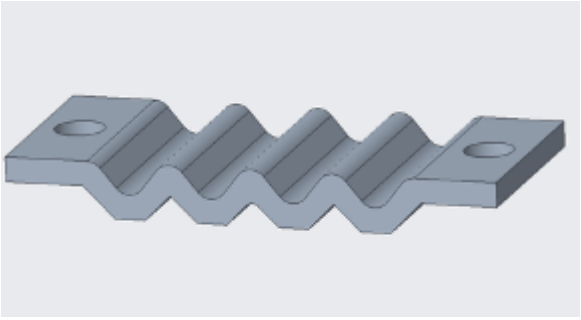
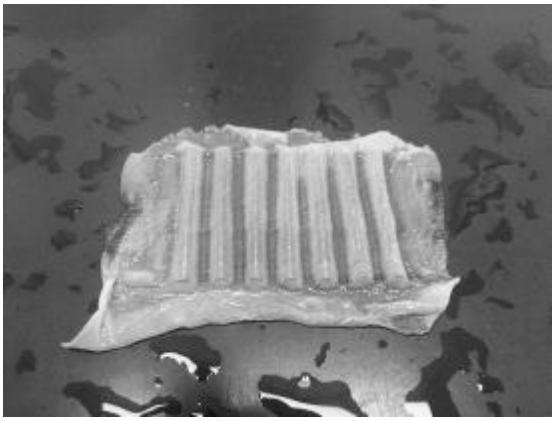


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P027

A modular in situ device for stress testing of biomaterials

*T. Dose[1,2], D. Krüger[1], J. U. Hammel[3], J. Moosmann[1], B. Zeller-Plumhoff[1], F. Wieland[1], I. Grewing[3], F. Beckmann[3], F. Wilde[3], R. Willumeit-Römer[1]
[1]Helmholtz-Zentrum Geesthacht, WB , Geesthacht, Deutschland
[2], ,, Deutschland
[3]Helmholtz-Zentrum Geesthacht, WPI , Hamburg, Deutschland

At the imaging beamline P05 [1] at the PETRA III storage ring at DESY, Hamburg, Germany a push/pull-out device was developed which allows the acquisition of tomographic data sets under static or cyclic load conditions. The design is modular such that the device can be adapted for different actuators and load cells in order to apply and monitor load over wide range of stresses. Device control and logging is fully integrated in the beamline control. Force closure between base plate and actuator is ensured by a cylindrical spacer made from PEEK or acrylic glass. Diameter and wall thickness of the spacers can be adapted to the envisage forces or experimental requirements. E.g. phase-contrast tomography benefits from large diameters of the spacer in order to avoid strong phase variations due to horizontally different optical path lengths. Dedicated 3D-printed sample mounts can be used to align and stabilize the sample. Contrast mechanisms available are conventional absorption contrast, grating interferometry, and propagation-based phase contrast. Future add-ons include monitoring and control of temperature and humidity.

Figure1: Set-up

Features of the modular stress system

- Wide range of stresses (~1 N … 1 kN)
- Spacer (wall thickness, diameter, material) adaptable to experimental requirements (force, contrast mechanism, etc)
- Flexible sample mount
- Option for additional sensors e.g. temperature, humidity
- Capability to rinse the cell with protective gas

Applications

- Morphological characterization of biodegradable metal implants [2, 3] in bone
- Characterization of tooth filling
- Studies of biomimetic materials
- Fatigue tests
- Investigation of metal-bone interfaces [4]
- Elucidation of implant degradation and failure mechanisms

First results

An implanted screw in a rat bone was suspended with different loads while tomograms were collected. The top row depicts a region of cortical bone. The bottom row depicts a region of the bone-to-implant interface.

Figure 2: Sequences of vertical slices through the tomographic reconstructions of an explant under increasing compressive load Julian Moosmann et al. unpublished

Acknowledgements

We would like to acknowledge support by the SynchroLoad project (05K16CGA) of the Röntgen-Ångstrøm-Cluster (RAC).

References

[1] F. Wilde et al., *AIP conference Proceedings* **1741** (2016), 030035.
[2] T. Dose et al., 5th Conference on Industrial Computed Tomography Proceedings, Shaker, Aachen 2014, page 421 - 424.
[3] T. Dose et al., 6th Conference on Industrial Computed Tomography (http://www.ndt.net/article/ctc2016/papers/ICT2016_paper_id51.pdf)
[4] T. Dose et al., Bionanomaterials Volume **15** Number S1, 2014, page S85

Figure 1

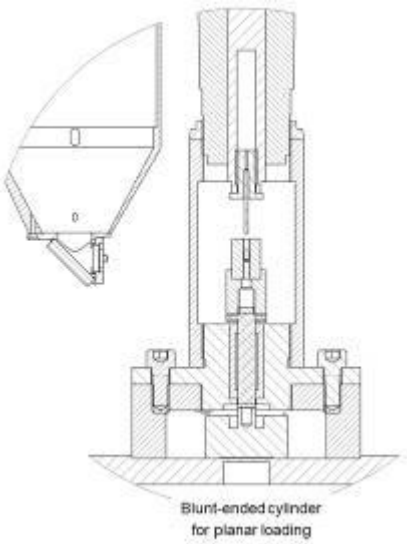
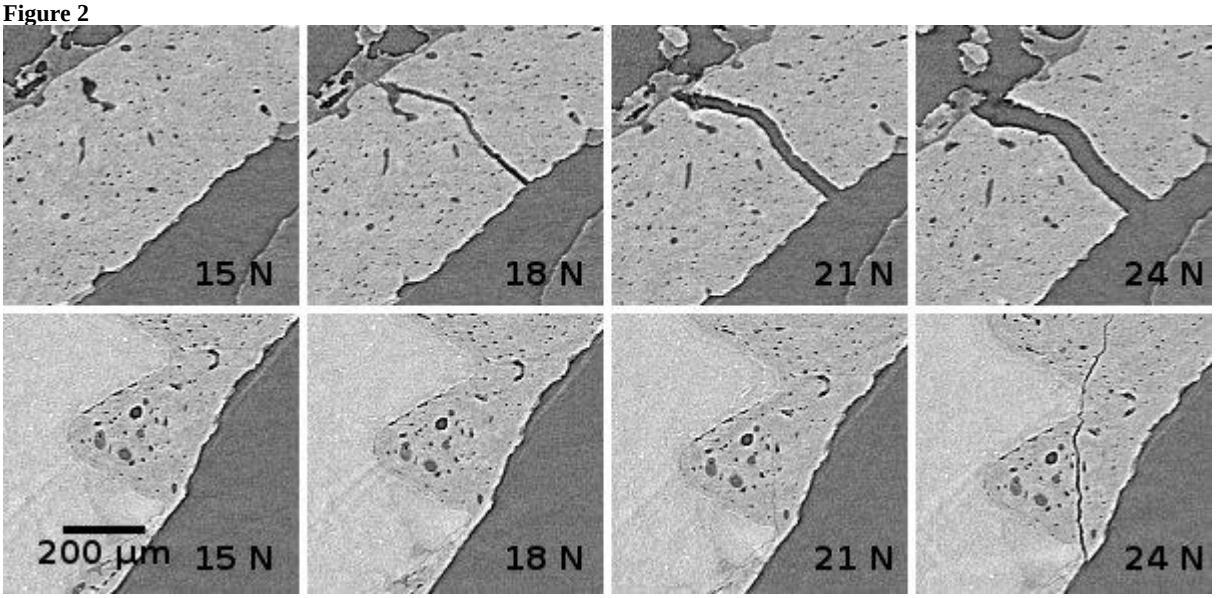


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P028

Biofunctionalization of polymeric implant material – impact of nanofibrous structure

*S. Kohse[1], K. Wulf[1], D. Bajer[1], N. Grabow[1], K. P. Schmitz[2], T. Eickner[1]
[1]Universitätsmedizin Rostock, Institut für Biomedizinische Technik , Rostock, Deutschland
[2]Institut für ImplantatTechnologie und Biomaterialien e.V., , Rostock, Deutschland

Minimization of neointimal hyperplasia with simultaneous promotion of the re-endothelialization of the polymeric surface is an important goal for successful cardiovascular application of polymer-based implants or implant coatings. Due to high hydrophobicity of the most synthetic polymers biofunctionalization of the implant surface is a common approach for improvement of cell-implant-interaction.

In the present study, vascular endothelial growth factor (VEGF) biofunctionalization of biocompatible polyurethane-co-silicone with different surface morphologies in combination with a simultaneous protein coupling and drug incorporation is investigated as a promising concept [1]. Therefore spraycoated films are compared to electrospun nonwoven material. As model drug for bulk incorporation fluorescein diacetate (FDAc) was used due to its diffusion and distribution coefficients similar to paclitaxel [2]. For evaluation, surface VEGF loading and release characteristics are determined and compared in dependence on surface morphology and FDAc drug loading. Furthermore, biocompatibility of functionalized samples are assessed by testing cell viability with L929 mouse fibroblasts.

In this study we present biofunctionalization of polyurethane-co-silicone surfaces. The increase of the surface to volume ratio is a major advantage of the fibrous structure resulting in a potentially higher surface loading. Furthermore preliminary studies indicate a synergistic effect of bulk incorporation and surface modification. So the use of polymeric nonwoven fabrics is a promising approach for cardiovascular applications.

Partial financial support by the Federal Ministry of Education and Research (BMBF) within RESPONSE "Partnership for Innovation in Implant Technology" and by the European Social Fund (ESF) within the excellence research program of the state Mecklenburg-Vorpommern Card-ii-Omics is gratefully acknowledged.

References

[1] Wulf K, Teske M, Löbler M, Luderer F, Schmitz K-P, Sternberg K. Surface functionalization of poly(ε-caprolactone) improves its biocompatibility as scaffold material for bioartificial vessel prostheses. Journal of biomedical materials research. Part B 2011; 98(1):89-100.

[2] Steele TWJ, Huang CL, Kumar S, Widjaja E, Chiang Boey FY, Loo JSC, Venkatraman SS. High-throughput screening of PLGA thin films utilizing hydrophobic fluorescent dyes for hydrophobic drug compounds. J Pharm Sci 2011;100:4317–4329.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P029

In vitro degradation experiments over one month period on magnesium-based alloys for biodegradable implant applications

*D. Krüger[1], B. Zeller-Plumhoff[1], J. Moosmann[1], F. Wieland[1], J. U. Hammel[1], R. Willumeit-Römer[1]
[1]Helmholtz-Zentrum-Geesthacht, , Hamburg, Deutschland

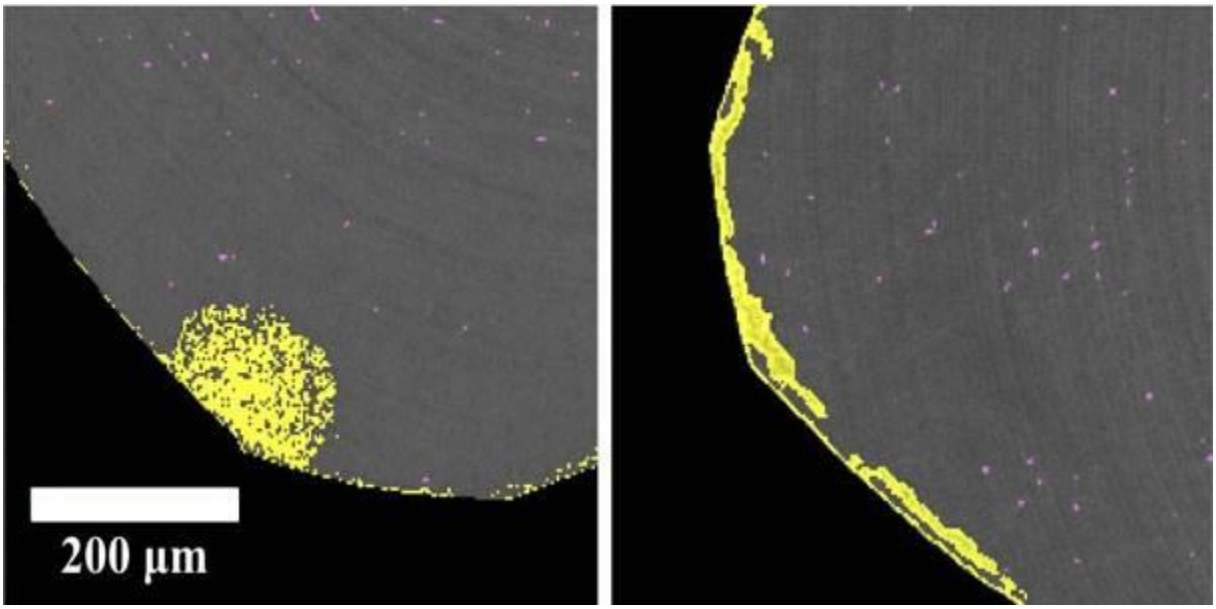
Magnesium (Mg)-based alloys can be applied as promising biodegradable implants due to their good biocompatibility, a *Young's modulus* similar to the one of bone and the ability for complete degradation [1]. The major advantage is the prevention of a second and costly removal surgery. This kind of application of Mg in medicine is only viable if the implant degradation takes place in a controlled and sufficiently slow manner [1]. To examine the degradation behaviour of two Mg-gadolinium (Gd) alloys with varying Gd content (5, 10 wt% Gd content) *in vitro* semi-static degradation tests over one month period were held. The samples had a shape of a screw (2x4 mm) and as degradation medium mimicking biological conditions α-MEM (Minimum Essential Medium) + 10% FBS (Fetal Bovine Serum) + 1% Pen Strep (Penicilin Streptomycin) was used. Investigations were performed using high-resolution synchrotron radiation computed tomography at beamline P05 at PETRA3 Deutsche Elektronen-Synchrotron (DESY), Hamburg. The analysis on the experimental data aim to extract the DR (degradation rate), DH (degradation homogeneity), dependency of Gd agglomerations on DR and DH. Also microstructural investigations were carried out using optical microscopy (OM), scanning electron microscopy (SEM) and electron backscatter diffraction (EBSD). First results indicate that Mg-5Gd exhibits bigger grain size (GS) (52.6±15.3 µm) than Mg-10Gd (GS = 25.9±0.1 µm). DH analysis show that Mg-5Gd screws tend to degrade with localized pitting corrosion in the first week of immersion. Afterwards the corrosion layer starts to spread over the whole surface. In contrast, the primarily localized pitting corrosion depth of Mg-10Gd is smaller, having a larger surface distribution (Fig. 1). An average DR = 0.30±0.09 mm/year for both alloys is observed over all 4 time points during one month period, which is acceptable for biodegradable implants [2].

Fig. 1: Exemplary cross sectional slices of screws after 1 week degradation: left) Mg-5Gd; right) Mg-10Gd. Yellow colored areas represent corrosion layers; magenta colored points represent Gd agglomerations.

References

[1] Regine Willumeit-Römer, Annelie Weinberg. 2015. Special issue "Biodegradable magnesium as implant material". 16(1):1-2.
[2] Olga W. Charyeva. 2015: Bioresorbable Magnesium Implants for Bone Applications. Justus Liebig University (Giessen).

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P030

Bioactive coating of polyetheretherketone (PEEK)

*T. Frankenberger[1], C. Ganz[1], T. Gerber[1]
[1]Universität Rostock, Institut für Physik, Nano- und Biomaterialien , Rostock, Deutschland

In addition to its favorable mechanical properties [1], PEEK is known to be inherent inert and does not chemically bind to soft and hard tissue as to no direct bone contact is promoted. For this reason, specimen were coated with a bioactive material and subsequently annealed to establish an interface consisting of PEEK and the biomaterial to enhance the bone growth on PEEK implants. The coating consists of a nanoporous silica matrix with embedded synthetic morphologic nanocrystalline hydroxyapatite (Si:HA = 39:61) [2]. Thermal annealing above the melting temperature transforms the solid PEEK surface in the range of viscous flow behavior leading to the ingression of molten PEEK into the interconnected nanopore structure of the coating. SEM and TEM images prove a successful merge between the two layers resulting in an interface-thickness of 1,65±0,27 µm. All interconnected micro and nanopores of the coating material within the interface are completely filled with PEEK. Consequently, the originally porous coating transformed into a dense interface through the ingress of molten PEEK. This micro mechanical connection built a favorable base for a well adhered coating. Electron Energy Loss Spectroscopy (EELS) of pure PEEK, HA and silica compared to the interface provides the evidence of PEEK being present in the interface area. *In vitro* investigations with human blood and calf serum within 48 h confirm the main mechanism of the grafting material, the exchange of the inorganic matrix into an organic one [2], as a coating and in the interface area. With increasing immersion time, the silica content in both, pure coating and interface were reduced. Compared to a coating of the thickness between 6 µm and 8 µm, the interface exhibits a delayed beginning and lower reduction speed of the silica. Cytotoxicity tests (XTT; L929) of pure PEEK, the interface and coating show a cell activity above 80 % confirming their nontoxicity. Live/Dead tests with fibroblasts (L929) of the same samples indicate only a few apoptotic cells and beginning cell spreading except at pure PEEK, leading to a high biocompatibility not only for the established biomaterial as a coating but also for the interface.

References

[1] S. Kurtz, J. N. Devine, PEEK biomaterials in trauma, orthopedic and spinal implants, *Biomaterials* 2007:28 4845-4869.
[2] M. Adam, In vivo and in vitro investigations of a nanostructured coating material – a preclinical study, , *International Journal of Nanomedicine*, 2014:9 975–984.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P031

Designing novel scaffolds for ocular surface reconstruction based on aligned nanofibers

*F. Küng[1,2], P. Stafiej[1,2], T. A. Fuchsluger[2], D. W. Schubert[3]
[1]Friedrich-Alexander-Universität Erlangen-Nürnberg, Lehstuhl für Polymerwerkstoffe , Erlangen, Deutschland
[2]Friedrich-Alexander-Universität Erlangen-Nürnber, Lehrstuhl für Augenheilkunde , Erlangen, Deutschland
[3]Friedrich-Alexander-Universität Erlangen-Nürnber, Bayerisches Polymerinstitut , Erlangen, Deutschland

Disease, chemical burns, cuts, and other incidents can cause damage to the most outer layer of the eye called the cornea. In order to cover these wounds and support the healing process ophthalmologists cover the damaged area with a patch made from the amniotic membrane, a piece of tissue harvested from the human placenta. Because of the varying properties of human tissue and the dependency on donors, there is a need for an artificial replacement for the amniotic membrane.

Based on this need we developed novel processing routes for building scaffolds from aligned nanofibers [1]. Aligned nanofibers were chosen because they offer high transparency so the patient"s vision won"t be disturbed too much and infections and other complications in the eye will be visible to the ophthalmologist. All nanofibers are produced using electrospinning including novel collectors to match the dome shape of the cornea (figure 1). In order to match the application conditions a novel testing procedure was developed [2] and performed to characterize the resistance against suture pull-out. Tests with human tissue were performed in order to benchmark and optimize the scaffolds (figure 2). As a further add-on the inclusion of hydrogels in the nanofiber matrix was investigated. Therefore this is shown to be a suitable novel approach for tissue engineering in ophthalmology.

References

[1] Schubert, Dirk W.; Stafiej, Piotr; Küng, Florian; Fuchsluger, Thomas A. (2016): Wound dressing compromising polymer fibers. In: *EU-Patent* 2016 application No. 16185130.8-1455.
[2] Küng, Florian; Schubert, Dirk W.; Stafiej, Piotr; Kruse, Friedrich E.; Fuchsluger, Thomas A. (2016): A novel suture retention test for scaffold strength characterization in ophthalmology. In: *Materials science & engineering. C, Materials for biological applications* 69, S. 941–946. DOI: 10.1016/j.msec.2016.07.052.

Figure 1

Figure 2

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P032

Transparency of suture able polycaprolactone nanofiber hydrogel compounds (NFHC) for medical application in ophthalmology

*P. Stafiej[1], F. Küng[1,2], T. A. Fuchsluger[2], D. W. Schubert[3,4]
[1]Friedrich-Alexander-Universität Erlangen-Nürnberg, Lehrstuhl für Polymerwerkstoffe , Erlangen, Deutschland
[2]Friedrich-Alexander-Universität Erlangen-Nürnberg, Lehrstuhl für Augenheilkunde , Erlangen, Deutschland
[3]Friedrich-Alexander-Universität Erlangen-Nürnberg, Bayerisches Polymerinstitut , Fürth, Deutschland
[4]Friedrich-Alexander-Universität Erlangen-Nürnberg, Bayerisches Polymerinstitut , Erlangen, Deutschland

The treatment of wounds, especially in ophthalmology, often requires donor tissue as replacement or wound closure material. Biological scaffolds (i.e. donor tissue) often have inconsistent mechanical and optical properties as well as insufficient availability. Therefore a strong focus was made on development of synthetic materials. Augmented replacements need to be able to resist the force applied on the scaffold during suturing while maintaining its transparency.

We developed scaffolds consisting of alginate hydrogel and compounded them with electrospun nanofibers made out of polycaprolactone and a blend with chitosan to reinforce the hydrogels.

The transparency of our scaffolds was quantified with a UV-Vis spectrophotometer over the visible light area ranging from 400 to 800 nm [1]. To visualize the transparency measurements were done using a method described by Schubert et. al. [2]. A line of text was printed out in different font sizes on which the scaffolds were placed to evaluate the readable font sizes of the text lines.

To examine the suture ability of the scaffolds a method developed by Küng et. al. [3] was used. A suture was passed through the sample in a distance of 1 mm to the rim before the sample was clamped in a tensile testing machine. While pulling out the suture the force load was measured which gives us the suture retention strength of the scaffolds.

Compounding both materials leads to scaffolds with high transparency (55-85 %) while maintaining enough mechanical strength for suturing the material in ophthalmic applications [4].

References

[1] Salehi S.; et.al.: Acta Biomater, 2017,50,370-380
[2] Schubert, DW; et. al.; Macromol. Symp., 2016, 365, 87-94
[3] Küng, F.; et. al.; Mater Sci Eng C, 2016, 69, 941-946
[4] Küng, F. et. al.; EU-Patent notifying number: 16185130.8-1455

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P033

Eluate and direct contact tests revealed promising biocompatible hydrogels of crosslinked ionic liquids for biomedical applications

*A. Brietzke[1], C. von der Ehe[2], S. Illner[1], C. Matschegewski[3], N. Grabow[1], U. Kragl[2]
[1]Institut für Biomedizinische Technik , , Rostock, Deutschland
[2]Universität Rostock, Institut für Chemie , Rostock, Deutschland
[3]Institut für Implantattechnologie und Biomaterialien e.V., , Rostock, Deutschland

For the development of intelligent implant systems hydrogels from crosslinked ionic liquids feature a high potential to be utilised as drug depot. Biocompatible materials with the ability to provide active ingredients can ensure stringent requirements on implants such as rapid wound healing and long-term functionality. While eluate testing methods analyse potential cytotoxic effects of material eluates on cell viability, direct contact testing proves the ability of cells to tolerate or even interact with materials. In this study the effect of different hydrogels and hybrid hydrogels on cell viability was investigated by both, in vitro eluate and direct contact tests.

Hydrogels were previously polymerised from a variety of different ionic monomers based on methacrylate, methacrylamide, styrene or vinyl imidazolium derivatives in aqueous solution. Within this free radical polymerisation process *N*,*N*′-methylenebisacrylamide was used as crosslinker. In order to prevent toxicity caused by residual monomers, these were subsequently thoroughly leached out of the hydrogels by several rinsing steps with deionised water. CellQuanti-Blue Cell Viability Assay Kit (BioAssay systems, Hayward, CA, USA) was implemented to proof cell viability.For eluate tests 2x104 L929 mouse fibroblasts (CCL-1, ATCC) were seeded in a 96-well microtiter plate with 200 µL culture medium per well and incubated under cell culture conditions (37 °C, 5% CO2) for 24 hours. Water expanded hydrogels were eluted in serum free culture medium under identical conditions. After elution, 10% FCS (fetal calf serum) was added to the sterile-filtered medium, and a dilution series was prepared. Subsequently, the cell culture media of the raised cells were exchanged with 200 µL of each eluate dilutions and incubated for 48 hours (37 °C, 5% CO2). For direct contact testing cells were directly seeded on the hydrogels and incubated for 48 hours under same conditions.

The predominant part of the hydrogel eluates generated only a marginal reduction of less than 15% cell viability at 100% eluate concentration.In prelimiary direct contact test no cytotoxic effects of hybrid hydrogels was found. This underlines the excellent suitability of these hydrogels for biomedical applications and revealed some promising candidates for the development of drug depots for implants. Future microscopic approaches will prove the suitability of the hydrogels to serve as a matrix for cellularisation.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P034

Re-Engineered Pericardium Sponges as Novel Biomaterials: Creation of Sponge-Like Tissue by Freeze Casting and Lyophilization

*T. Igelspacher[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Collagen sponges are commercially available and are used in clinical practice for dental repair [1], as wound dressing for burn wounds [2] and other applications. These collagen sponges are mainly fabricated from acid-insoluble, swollen collagen derived from bovine or porcine tendon and skin. Porcine pericardium is used for over 30 years for biological heart valve prosthesis [3]. Due to its composition, pericardial tissue can be used as a collagen source. Instead of using an elaborate purification process, we use the pericardium in its entirety and thus preserve its native properties.

Prior to homogenizing the porcine pericardium (PP) is obtained from the slaughter-house and freed from undesired remnants. The mechanical pulping of the PP is done by using a tissue homogenizer, where small ceramic beads (ZrO2) are highly accelerated by the 3D-movement of a tube that is filled with the ZrO2 beads, the PP and a fluid, e.g. 0.9% NaCl solution. The tissue is broken up by the impact of the spheres. To ensure that during homogenization the collagen does not denature, the device is cooled by a cryo unit using a stream of compressed air precooled by liquid nitrogen. Afterwards the resulting tissue suspension is filtrated to obtain a defined particle size range.

The pericardial suspension is frozen in predefined molds at < -25°C in a freezer. This ice-casting step creates a porous structure and leads to the formation of a sponge-like tissue structure. After about 6h the frozen pericardial material is lyophilized for about 24h. Lyophilized scaffolds are cross-linked for 24h in 0.5% GA at 4°C and afterwards in 0.5% GA for 1h at 60°C.

The resulting re-engineered pericardium sponges are tested for tensile and compression properties, resistance to enzymatic degradation and its pore structure.

References

[1] S. Jimson et al. J Pharm Bioallied Sci. 2015 Apr; 7(Suppl 1): S134-S137.
[2] C. H. Lee et al. International Journal of Pharmaceutics 221 (2001) 1–22.
[3] F. J. Schoen et al. CCC 0021-9304/99/040439-27.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P035

The Suture Retention Strength of Bacterial Cellulose

*T. Pieger[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Bacterial cellulose is a linear-chain polymer with interesting structure and properties for medical applications. Due to its high purity (free of lignin, hemicellulose and pectin) and water content (more than 90%) [1] it can be used in cosmetics, as artificial skin or blood vessels as well as for wound healing [2,3].

Especially the application as artificial blood vessels requires good mechanical properties and a low roughness of the inner surface [3]. The material must be easy to sew to human blood vessels and the suture must not tear under load. To analyze this feature the suture retention strength is defined.

The suture retention test is performed as a uniaxial tensile test. A suture loop is applied to one side of the specimen with a surgical needle at a distance of 1 mm from the edge. The suture is wrapped around a screw and fixed between shims. The other end of the specimen is fixed in a clamping jaw. The sample preparation and the uniaxial tensile test have to be performed under wet conditions in water to avoid drying of the material. The thickness of the sample material was measured with a caliper. Each specimen is used only once.

The suture retention strength is calculated from three parameters: the thickness of the sample, the diameter of the suture and the maximum force at break. It is given by the following equation in N/mm^2: [4]

suture retention strength = force at break/(sample thickness x suture diameter)

In general, the suture retention strength of the bacterial cellulose depends on the bacterial strain, the culture conditions and the sample preparation. In the latter case the edges are essential. They should be cut cleanly. Furthermore the thickness of the material should be constant.

Results of elaborated tests of cellulose prepared under different culture conditions will be presented.

References

[1] D. Klemm et al, Angew. Chem. Int. Ed., 2005, 44, 3358-3393
[2] P. R. Chawla et al, Food Technol. Biotechnol., 2009, 47(2), 107-124
[3] D. Klemm et al, Prog. Polym. Sci., 2001, 26, 1561-1603
[4] F. J. Obermiller et al, Med. Sci. Monit, 2004, 10(1), 1-5

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P036

Creation of Sponge-Like Tissue from Homogenized Porcine Pericardium by a Modified Freeze Casting Technique

*T. Igelspacher[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

A commonly used biological tissue for cardiovascular implants is pericardium [1], either from bovine or porcine source. Pericardial tissue is mainly composed of collagen, mostly type I, and other constituents like elastin, glycosaminoglycans and proteoglycans [2]. With its high collagen content, pericardium can be used as a collagen source for a wide range of applications that must not be restricted to cardiovascular implants only. By fragmenting the pericardium to small units, homogenizing them and reassembling them by chemical cross-linking a novel material is created, the re-engineered pericardium sponge.

Prior to homogenizing the native porcine pericardium which is obtained from the slaughter-house it is thoroughly cleaned and undesired remnants are removed. The mechanical pulping of the pericardium is done by using a tissue homogenizer. Small ceramic beads (ZrO2) are highly accelerated by the 3D-movement of a tube that is filled with the ZrO2 beads, the pericardium and a fluid, e.g. 0.9% NaCl solution. The tissue is fragmented by the impact of the spheres. To ensure that during homogenization the collagen does not denature, the device is cooled by a cryo-unit using a stream of compressed air precooled by liquid nitrogen. The resulting tissue suspension is filtrated to obtain a defined particle size range.

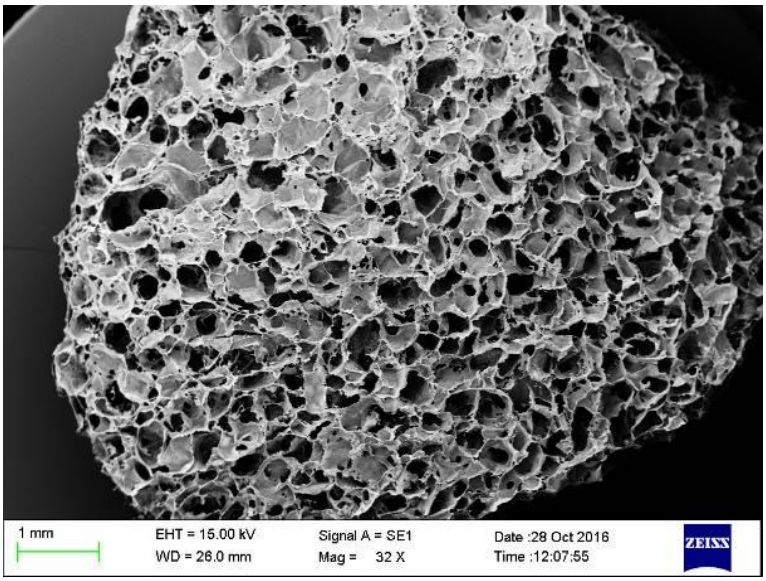
In order to create a sponge-like tissue from the homogenized suspension a modified freeze casting technique is used. A solution of glutaraldehyde (GA, 0.5%) is deep-frozen as a mold with the negative shape of the desired component. In this way almost any 3D-shape can be obtained. The pericardial suspension is then injected into the mold and kept in the freezer until it is completely frozen. The advancing freezing front in the suspension pushes the fragments of the pericardium ahead and thus leads to the highly porous structure of the sponge-like tissue that is finally obtained. During thawing of the assembly the sponge is cross-linked by the GA that is set free from the mold. To stabilize the structure further additional cross-linking at higher temperatures (e.g. 60°C) can be applied.

The resulting sponge-like tissues are tested for tensile and compression properties, resistance to enzymatic degradation and pore structure (Fig. 1).

References

[1]E. Rémi et al. In: Biomaterials Science and Engineering: In Tech (2011), S. 437–455.
[2] A. S. Braga-Vilela et al. J Membrane Biol (2008) 221:15–25

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P037

Chemical Bonding of Patches of Porcine Pericardium by Glutaraldehyde-Fixation

*D. Niopek[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Porcine pericardium is a biomaterial used for biological implants, including the construction of a variety of bioprostheses such as vascular grafts and heart valves. [1] As a crosslinking agent glutaraldehyde (GA) is commonly used to ensure the chemical long-term stability of collagen-based biomaterials and to lower the immune reaction after implantation. Although a large variety of chemical pathways may be involved in this crosslinking, it is known, that GA reacts primarily with amino groups of proteins. [2, 3] The aim of this work is to achieve chemical bonding between patches of pericardial tissue by GA-crosslinking.

For this purpose native porcine pericardium was cleaned and then cut into rectangular strips of 30 mm x 5 mm. Two strips were placed between acrylic plates with an overlap of 5 mm x 5 mm. The mounting of the acrylic plates was realized by screws using a torque spanner at 0.5 Nm to ensure a reproducible stress condition. The samples were crosslinked in 0.5 % glutaraldehyde (50 % stock solution diluted in DPBS) for 6 days. Additionally a control group, stored in pure DPBS, was prepared in order to verify the effect of glutaraldehyde on the interconnection.

In contrast to the control group, a stable connection between two tissue strips could be established by crosslinking with glutaraldehyde. The glutaraldehyde not only creates chemical bonds within a single pericardium patch, but also between two overlapping patches. Performing uniaxial tensile testing, the stability of this connection was analyzed. An average fracture force of 3.87 N ± 1.4 N was observed.

Crosslinking with glutaraldehyde leads to chemical bonding of native porcine pericardium patches. Additional research is required to determine the ideal parameters to achieve a solid tissue connection, including GA-concentration, fixation temperature and time as well as area and geometry of overlap.

References

[1] Rémi et al. *Biomaterials Science and Engineering 437-456*
[2] Damink et al. *Journal of Materials Science: Materials in Medicine 6 460-472*
[3] Cheung et al. *Connective Tissue Research 10 187-199*

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P038

Setup for Testing the Compression and Swelling Properties of Sponge-Like Biomaterials

*T. Igelspacher[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Sponge-like biomaterials, e.g. a tissue matrix of homogenized porcine pericardium, offer a wide range of applications in medical implant technology. Two main characteristics of any sponge-like material are the compression and swelling properties and the closely related pore structure.

We present a setup to test the compression resistance and the temporal swelling of sponge-like materials. While the compression resistance can be tested in the dry and the wet state, swelling only occurs in the presence of a liquid.

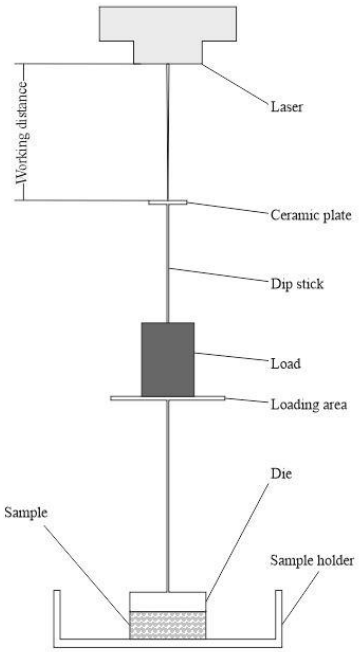
Basically, there are two possible modes of operation of such type of setup. Either, a defined indention force (weight) is applied and the indention is measured or a defined indention is applied and the indention force is measured. We utilize the first mode in our setup.

Fig. 1 shows a basic drawing of the test setup. The laser-assisted distance sensor on top measures the distance to the dip stick. The fully guided dip stick moves up during swelling of the specimen and down when the sample is compressed. On the upper end there is a ceramic plate for better laser detection and at the bottom there is a die. Additionally, a loading area is connected to the dip stick at half height where a defined load can be placed manually.

The compression resistance is measured as follows: in order to record the starting thickness of the sample, the dip stick is placed on the sample without any additional indention load for 10s and the initial thickness dA is recorded. Subsequently a predefined indention load is applied to the loading area and the sample is allowed to settle for 60s before a second thickness value dB is taken. The relative decrease in thickness of the sample ((dA/dB) * 100%) is defined as its compression resistance against vertical load.

To measure the swelling behavior of a sponge-like material, the sample is dried and pressed to a defined thickness. After placing the dry sample under the die and taking the initial thickness, a fluid, e.g. NaCl solution, is filled in the sample holder to start the swelling process. The thickness as a function of time is detected with a frequency of 10Hz and visualized in a graph.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P039

3D Tissue Forming of Porcine Pericardium by Glutaraldehyde-Fixation using Porous PLA-Molds

*D. Niopek[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander-Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Porcine pericardium is a biomaterial commonly used for biological implants, including the construction of a variety of bioprostheses such as vascular grafts or heart valves. [1] The chemical fixation with glutaraldehyde (GA) ensures the long-term durability of collagen-based materials and reduces an immune reaction after implantation. [2] The general objective of this work is to use the crosslinking process with glutaraldehyde not only for chemical stabilization of the pericardium, but also for 3D tissue shaping.

For this aim the porcine pericardium is inserted in 3D printed molds, manufactured from polylactic acid (PLA) and then treated with 0.65 % GA-solution (50 % stock solution diluted in DPBS) for 6 days. The porosity of the PLA-molds allows the supply of glutaraldehyde solution and thus ensures a homogenous tissue fixation.

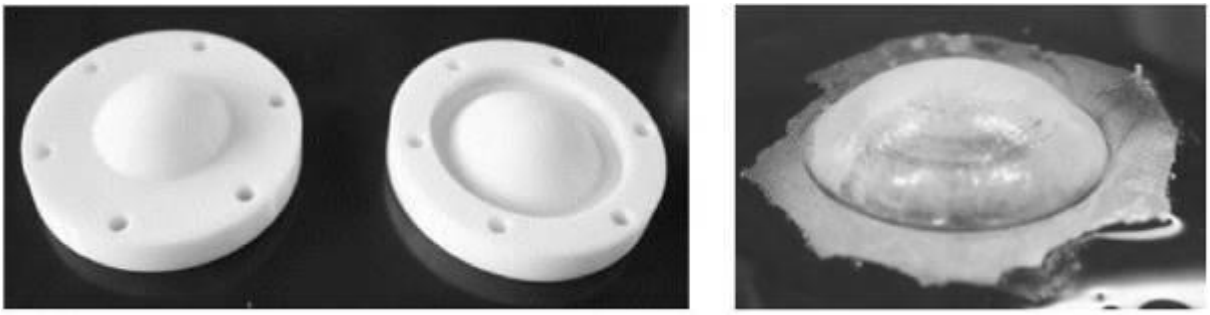
During the fixation process the form of the 3D molds is transferred to the tissue. After removal of the tissue from the molds, the 3D form is stable. It is thus possible to obtain almost any desired 3D tissue shape, e.g. spheres, edges, waves and spikes. In addition it has been shown that this technique of tissue forming also works on a microscopic scale as even smallest irregularities or engravings on the surface of the molds can be reproduced on the tissue. In figure 1 a spherical mold with counterpart (left) and pericardial tissue after crosslinking with GA (right) is illustrated.

It can be concluded that crosslinking with glutaraldehyde enables a permanent 3D shaping of porcine pericardium. Thus, it is possible to accurately adapt the geometry of the tissue to its application. Further research is needed to analyze the influence of the GA-concentration, fixation temperature, fixation time and initial load on the strength of shape of the tissue.

References

[1] Rémi et al. *Biomaterials Science and Engineering 437-456*
[2] Gavilanes et al. *Connective Tissue Research 13 37-44*

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P040

Bacterial Cellulose as a Novel Biomaterial

*T. Pieger[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Cellulose is a linear-chain polymer composed of several linked β-1,4-glucans and it is the most common organic polymer [1,2]. It can be used for wound healing, as artificial skin or as artificial blood vessels. [2,3]

Apart from plants, there are certain bacteria that produce cellulose such as Acetobacter, Aerobacter and Achromobacter strains. Compared to plant cellulose the bacterial cellulose is of high purity (free of lignin, hemicellulose and pectin) and has a water content of more than 90%. Its degree of polymerization is 2000-8000 and the crystallinity shows values of 60-90%. After air drying the cellulose shrinks and regains a water content of only 6% after rehydration. This value is near to that of plant cellulose, but can be increased to 70% by using freeze-drying as drying process. [1]

The most well analyzed and effective cellulose producers are Acetobacter xylinum and Acetobacter hansenii [1,2]. They synthesize the cellulose at the interface between air and a culture medium containing D-glucose. The glucose is transformed to uridine diphosphate glucose (UDP glucose) and added to the end of the polymer chain by cellulose synthesized complexes (terminal complexes). The chains exit the bacteria cells through the pores located at the cell surface as elementary fibrils where they aggregate to microfibrils. The microfibrils are assembled to form ribbons building a 3D network**.** [1,2,3]

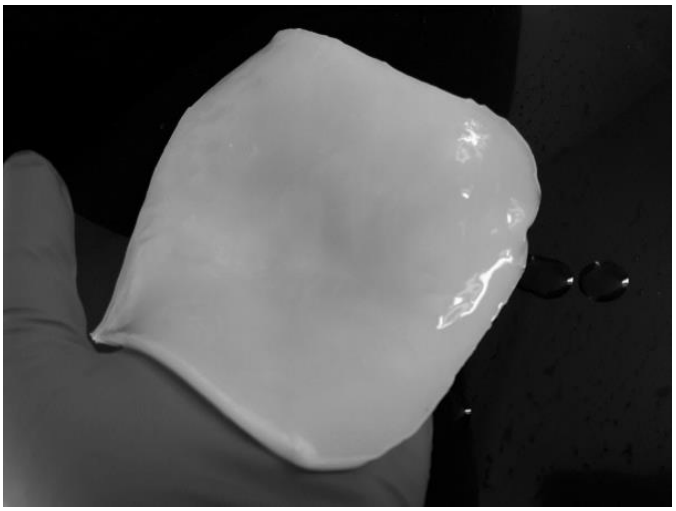
For the cultivation and production of the bacterial cellulose a liquid medium is needed. The most known medium is that of Hestrin and Schramm [4]. By varying its composition, the cultivation time, the temperature and the pH value, it is possible to change the mechanical properties as well as the thickness of the cellulose. The selection of the bacterial strain also plays a major role. While Acetobacter strains are gram negative and not pathogenic [1], Sarcina ventriculi are gram positive [2]. However, both strains require oxygen for the formation of cellulose [1,2].

In figure 1 the bacterial cellulose produced by Acetobacter xylinum is illustrated.

References

[1] D. Klemm et al, Angew. Chem. Int. Ed., 2005, 44, 3358-3393
[2] P. R. Chawla et al, Food Technol. Biotechnol., 2009, 47, 107-124
[3] D. Klemm et al, Prog. Polym. Sci., 2001, 26, 1561-1603
[4] S. Hestrin and M. Schramm, Biochem. J., 1954, 58, 345-352

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P041

Effect of Yeast Extract on the Growth of Bacterial Cellulose

S. Buchholz[1], J. Völkel[1], *T. Pieger[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Bacterial cellulose (BC) is a biopolymer with outstanding structural features and excellent physio-chemical properties. Not only its high crystallinity but also its high mechanical strength, as well as its biocompatibility makes it interesting for various biomedical applications. Due to its great potential it is already used as artificial blood vessels, wound dressings, scaffolds for tissue engineering or as artificial skin, e.g. for burn treatment or skin regeneration. [1]

Different groups already analyzed the impact of carbon sources on cellulose production [2]. Here the influence of yeast on the yield of cellulose is shown.

Acetobacter xylinum (A. xylinum) is used for the production of BC with a culture medium by Hestrin and Schramm having a 30 % higher amount of citric acid [3]. To determine the influence of different amounts of yeast extract on cellulose production, two different series of experiments are prepared: one standard medium (A) and one medium with a higher amount of yeast extract (B). The cultivation medium is inoculated 1:16 with A. xylinum.

The 7-day cultivation takes place under aerobic static conditions in an incubator offering a temperature of 28° C. After the incubation period, BC is formed as a white thick layer on the surface of the medium. This layer is lifted from the medium and washed thoroughly in pyrogen-free water. To determine the yield of cellulose, the thickness is measured.

Comparing the two experimental series a 53.10 % higher yield of cellulose was noticed at series B. This shows that a higher amount of yeast extract significantly increases the yield of cellulose. Temperature, pH value and oxygen content can have a big impact on cellulose production as well. Changing the other ingredients of the culture medium, such as glucose, peptone or citric acid, also influences the volume of synthesized BC. Another option, which is used in current research, is to add Carboxymethylcellulose (CMC) or ethanol to maximize the amount of BC. [4]

References

[1] N. Shah et al., Carbohydrate polymers 98 (2) (2013) 1585–1598
[2] Dianne R. Ruka et al.,Carbohydrate polymers 89 (2012) 613-622
[3] Hesterin S. et al., Biochem J 58 (2) (1954) 345-352
[4] Gromet-Elhanan Z. et al., J Bacteriol 85 (1963) 284-92

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P042

Formation of Artificial Blood Vessels using Bacterial Cellulose and Characterization of Mechanical Properties

J. Erath[1], *T. Pieger[1], A. Rzany[1], B. Hensel[1]
[1]Max Schaldach-Stiftungsprofessur für Biomedizinische Technik, Friedrich-Alexander Universität Erlangen-Nürnberg, , Erlangen, Deutschland

With its fine nanofiber network Bacterial Cellulose (BC) is a very promising biomaterial. It is possible to form BC in individual tubular shapes [1] and it can be very attractive for vascular surgery, creating artificial blood vessels with different size, especially in the form of small diameter vascular grafts [2, 3]. We present the formation of these tubes and the characterization of their mechanical properties.

Generally BC can be formed at the interface between culture medium and oxygen. We offer two methods for the production of the tubes. For both methods we use an oxygen permeable silicone tube with an inner diameter of 9 mm and an outer diameter of 11 mm. In the first approach cellulose is formed inside the tube. In the second approach the cellulose is produced on the outer surface of the tube. The proportion of the bacteria Acetobacter xylinum and the culture medium is the same for both methods. For the characterization of these cellulose tubes wall thickness measurements were performed. For evaluating their mechanical properties the tensile force of the tubes was measured.

The dense and smooth cellulose tubes at the surface of the silicone tubes are formed during one week. The first approach yields thicker tubes (wall thickness around 0.35 mm) than the second approach (0.20 mm). The tubes with the thicker cellulose typically showed better mechanical properties (tensile force: inside: 6-8 N; outside 3-4 N). The first method yields thicker tubes as more oxygen is interacting with the culture medium. Generally it can be demonstrated that there is a correlation between the thickness of the tubes and their mechanical properties. The second approach has the advantage that the inner diameter of the created tubes is known which is very important for flow condition in artificial blood vessel.

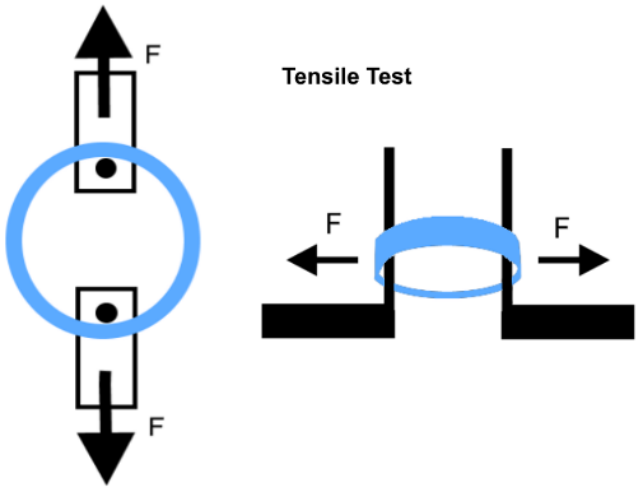
We present first results obtained for tubes produced from bacterial cellulose. They show good mechanical properties and offer a great potential for the vascular surgery.

In figure 1 the tensile test of the cellulose tubes for the characterization of the mechanical properties is illustrated.

References

[1] H. Bäckdahl et al. *Materials Science and Engineering C* 31 (2011); (1): 14-21
[2] D. Klemm et al. *Prog. Polym; Sci*. 26 (2001): 1561–1603.
[3] H. Bäckdahl et al. DOI 10.1002/bit.21314

References 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P043

Hydrogelation - Rheological aspects in polymerization of hybrid hydrogels

*S. Illner[1], O. Sahmel[1], S. Siewert[2], T. Eickner[1], N. Grabow[1]
[1]Institut für Biomedizinische Technik, Universitätsmedizin Rostock , Rostock, Deutschland
[2]Institut für Implantattechnologie und Biomaterialien e.V., , Rostock, Deutschland

Development of new implant coatings with stimulus-responsive drug release to treat infections after device implantation can be triggered by highly elastic hydrogels with adequate stability and adhesive strength in the swollen state. Examples of an ionic liquid ($[ViPrIm]^+[Br]^-$) as additive to poly(N-isopropylacrylamide) (pNIPAAm) showed unique effects on volumetric changes and mechanical properties as well as thermoresponsive drug release.[1]

In this context, rheological measurements allow studies involving the monitoring of hydrogelation processes as well as chemical, mechanical, and thermal treatments and effects of additives.[2] Various hybrid hydrogels were prepared by radical emulsion polymerization with or without 3D-crosslinking agent. Rheological properties of the multi-compound system during polymerization varying monomer, initiator and crosslinker amounts were monitored over time. For oscillatory time sweep experiments a RheoStress 1 (Thermo Haake GmbH, Karlsruhe, Germany) with a cone-plate C 20/1° Ti system was used.

The time dependence of the storage modulus (G') and the loss modulus (G") was measured, whereby the intersection of G' and G" indicates the sol-gel transition. Remarkable results regarding viscoelastic behavior of crosslinked and non-crosslinked hydrogels and complex viscosity of the formed hybrid hydrogels were obtained. For further material characterization rheology can be used to analyze the gelation determining processing capability and optimal working conditions. By the use of this method complete polymerization interconnecting all compounds and uniform gelation process for biomedical applications can be received providing the possibility to process mechanically stable, thermosensitive swellable implant coatings or wound closures.

References

[1] Pfensig S; Arbeiter D; Schmitz KP; Grabow N; Eickner T; Illner S (2016): Synthesis and characterization of PIL/pNIPAAm hybrid hydrogels. *CDBME*. 2(1): 1–4 DOI 10.1515/cdbme-2016-0004.
[2] Zuidema JM, Rivet CJ, Gilbert RJ, Morrison FA (2014): A protocol for rheological characterization of hydrogels for tissue engineering strategies. *J Biomed Mater Res B Appl Biomater*. 102: 1063–1073 DOI 10.1002/jbm.b.33088.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P044

Preventing implant-associated infections using a natural, biodegradable, antibacterial coating

J. Reinbold[1], A. K. Uhde[1], I. Müller[2], C. Schlensak[1], H. P. Wendel[1], *S. Krajewski[1]
[1]University Hospital Tuebingen, Department of Thoracic, Cardiac and Vascular Surgery , Tuebingen, Deutschland
[2]Albstadt-Sigmaringen University, , Sigmaringen, Deutschland

Implant-associated infections are one of the most serious complications after surgical interventions. Implants such as vascular grafts, hernia meshes, and surgical suture materials are optimal surfaces for the adherence of bacteria and subsequent colonization and biofilm formation. Due to a significant increase in antibiotic-resistant bacterial strains, naturally occurring agents exhibiting antibacterial properties have great potential in prophylactic therapies.

Hence, the aim of our study is the development of an implant coating consisting of PLGA as a biodegradable drug delivery system and the antibacterial substance Totarol, a naturally occurring diterpenoid isolated from *Podocarpus ssp.* Totarol was purified using column chromatography, and its antibacterial properties on *Staphylococcus aureus* strain RN6390 were shown using various bacterial analytical tests. Different implants, such as meshes, vascular grafts and sutures, were coated with a mixture of Totarol and PLGA, and the antibacterial properties were verified at different times *in vitro* and *in vivo*. In addition, hemocompatibility and cytotoxicity investigations showed that the Totarol-based coating exhibited no adverse effects.

Overall, the data indicates that our innovative implant coating has the potential to efficiently inhibit postoperative implant infections over a certain period without causing cytotoxic effects.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P045

Roles of microstructured PDMS surfaces with different geometries in promoting adipose-derived stem cell functions and nuclear distribution

*S. Nellinger[1], M. Schneider[1], R. Kemkemer[1,2], P. J. Kluger[1,3]
[1]Hochschule Reutlingen, , Reutlingen, Deutschland
[2]Max Planck Institute for Intelligent Systems, , Stuttgart, Deutschland
[3]Fraunhofer Institute for Interfacial Engineering and Biotechnology, , Stuttgart, Deutschland

The control of stem cell function by surface topography is a promising tool regarding diverse medical applications including implatable medical devices and scaffolds for tissue engineering. The effects of surface geometry on adipose-derived stem cell (ASC) behavoir are highlighted by many studies. It is well known that cells interact at a high level with their surrounding extracellular matrix (ECM) *in vivo* and growth substrates or scaffolds *in vitro*. The surrounding material can provide cues that guide cell behavoir. In the context of cell based therapies it is essential to understand how material surfaces influence stem cell behavior. In the present study the influence of microstructured polydimethylsiloxane (PDMS) surfaces with different dimensions and geometries on ASC functions was examined. ASCs were isolated from tissue samples from donors undergoning plastic surgery by enzymatic digestion. PDMS molds with different microstructures were produced using softlithography. Contact angle meassurment of PDMS molds revealed a higher hydrophilicity after O2 plasma treatment. ASCs were cultured on these surfaces for 21 days in growth medium. It was found that ASCs adhere onto different microstructures of PDMS. Orientation of ASCs on the different microstructures was proven by phalloidin staining of F-actin and focal adhesion distribution was proven by immunofluorescenc staining of the focal adhesion protein vinculin. After 21 day culture period ASCs exhibit a structure related orientation depending on microtopographic dimension and geometry. Further it can be observed that ASCs cultured on microstructured surfaces exhibit longer pseudopodia compared to the planar control. Nuclear distribution of ASCs was proven by DAPI staining. It was found that nuclear distribution correlates with microstructure dimension and geometry. Proliferation of ASCs was determined on the different microstructures. Oil red O and alizarin red staining revealed no adipogenic or osteogenic differentiation. To further investigate the influence of microstructure of surrounding material on ASCs different geometries and chemical and physical modifications should be examined to their effect on ASC functions and nuclear distribution.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P046

In vivo study on strontium enriched calcium phosphate bone cements with and without mesoporous bioactive glass

*C. Kern[1], N. Döhner[2,3], S. Ray[3], L. Franz-Forsthoffer[3], I. Kramer[3], U. Thormann[3], V. Alt[3], M. Schumacher[4], M. Gelinsky[4], M. Rohnke[2], J. Janek[2]
[1]Justus-Liebig-Universität Gießen, Physikalisch-Chemisches Institut , Gießen, Deutschland
[2]Justus-Liebig-Universität Gießen, Physikalisch-Chemisches Institut , Gießen, Deutschland
[3]Justus-Liebig-Universität Gießen, Labor für Experimentelle Unfallchirurgie , Gießen, Deutschland
[4]Technische Universität Dresden, Zentrum für Translationale Knochen-, Gelenk- und Weichgewebeforschung , Dresden, Deutschland

Osteoporosis is a systemic bone disease, characterized by reduction in bone density resulting in an increased risk of bone fracture. Integration of orthopaedic implants, e.g calcium phosphate (CaP) bone cements, and regeneration of damaged bone is crucial for long-term clinical success. CaP cements have a chemical similarity to the natural bone mineral phase, possess excellent biocompatibility, are resorbable and osteoconductive. Osseointegration of these implants and enhancement of bone defect healing can be locally stimulated by incorporation of therapeutic substances, e.g. Sr^{2+}. Numerous studies have shown that Sr^{2+} has a dual effect on bone metabolism: it both stimulates osteoblastic bone formation and reduces osteoclastic bone resorption.

The first studied bone cement is a Sr^{2+} enriched CaP cement paste containing a biocompatible oil. Hardening of the cement starts only after implantation. Therefore, the paste can easily be applied into any bone defect. The second cement paste is a derivative of the first modified with 10 wt-% of mesoporous bioactive glass particles (MBG) which are embedded into the CaP bone cement matrix. After implantation the glass particles is expected to create pores in the material due to a higher solubility than the cement matrix. This induction of porosity could help to enhance bone tissue ingrowth of new bone and osseointegration of the cement.

Bone samples were obtained 6 weeks after cement implantation in critical-size metaphyseal fracture defects in the femur of osteoporotic rats. Release of Sr^{2+}, calcium and collagen mass distribution of bone cross sections with incorporated biomaterial are studied by time of flight secondary ion mass spectrometry (ToF-SIMS). Histological analysis and histomorphometry were performed using Movat and Von Kossa/van Gieson staining to assess new bone formation within the former defect area.

Sr^{2+} is detected in bone sections by ToF-SIMS analysis in bone cement remnants, biomaterial/tissue interface regions and up to 4-6 mm from the implant area. This demonstrates the successful *in vivo* release of Sr^{2+} from both bone cements into bone. Fragmentation of biomaterials was shown histologically and in ToF-SIMS analysis with higher degradation of the bone cement containing MBG. Glass pores inside the remaining MBG containing cement could be detected via ToF-SIMS. Histology revealed increased amount of new bone formation, chondrocyte activity and osteoid formation in the MBG containing biomaterial.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P047

Development of a chip-platform for investigating cellular response to surface curvature

*K. Frey[1], M. Schneider[1], K. Athanasopulu[1], R. Krastev[1,2], R. Kemkemer[1,3]
[1]Hochschule Reutlingen, , Reutlingen, Deutschland
[2]NMI, Natural and Medical Sciences Insitute at the University of Tübingen, , Reutlingen, Deutschland
[3]Max-Planck-Institute for Medical Research, , Heidelberg, Deutschland

Surface topographies are often discussed as parameter influencing cellular behavior like orientation or directed migration. Whereas most *in vitro* studies use surfaces with micro-fabricated structures with sharp edges, smoothly curved topographies on the micrometer-scale might be more relevant in mimicking the native cellular environment. Structures with curved surfaces in different dimensions, like blood or lymphatic vessels, osteons or collagen fibers are omnipresent in situ.

Cellular responses and the underlying mechanism of curvature sensation are not yet fully understood. Theoretical studies[1] predict morphological adaption of cells to such curved surface features suggesting that the cell optimizes their active contractility, and the anisotropic, bending response of the stress fibers induced by the curvature, resulting in an angle of orientation of the cell on the curved surface.

However, systematic experimental studies are rare but would help to reveal the molecular mechanism of curvature sensing.

Therefore, we developed a chip-platform with systematically varied curvatures on a surface by using photolithographic methods with photoresist thermal reflow and soft lithography in order to study the effect of different radii of curvatures on the cell structure and functional responses.

An initial aspect ratio of 1:3 (height to width) leads to either semi-spherical or semi-cylindrical structures in arrays with extension of several mm (Figure 1). Subsequent repetitive molding enables us to produce both convex and concave surface topographies with curvature radii varying from approximately 5µm to 75µm, displaying the different dimensions of curvatures that are expected to have influence on cellular behavior.

Our platform facilitates the study of both, the general impact of substrate curvature and of the resulting change in cytoskeletal forces, as well as the influence of (pharmacological) manipulation of the cytoskeleton rigidity on the orientation angle of the cell.

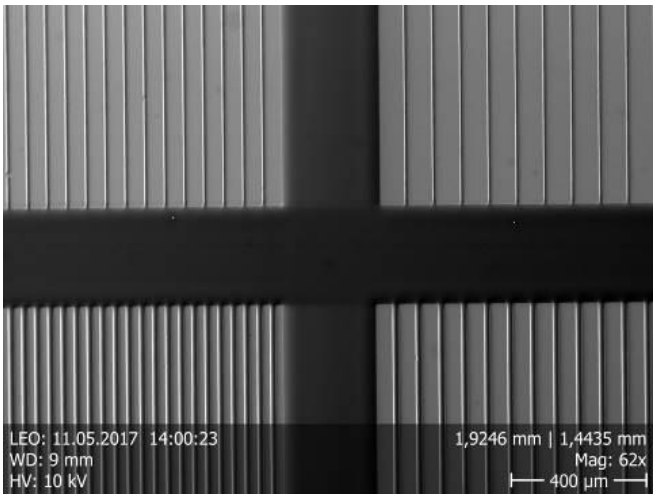
This will not only help understanding the mechanism of curvature sensation but the results will also contribute to the optimization of cell-biomaterial interaction.

References

[1] Y.Y. Biton, S.A. Safran, Physical Biology 2009, 6, 46010

Figure 1: SEM image of cylidrical surface structures with varying dimensions.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P048

Nanoporous Platinum Coatings for Neural Interface Applications

*K. D. Kreisköther[1], H. C. Schwarz[1], N. Ehlert[1], P. Behrens[1], J. Schulze[2], A. Warnecke[2]
[1]Leibniz Universität Hannover, Institut für Anorganische Chemie , Hannover, Deutschland
[2]Medizinische Hochschule Hannover, Klinik für Hals-Nasen-Ohrenheilkunde , Hannover, Deutschland

By enhancing the long-term biointegration and the contact between electrode and nerve fibers, the function of neural interface electrodes, e.g. cochlear electrode, can be improved. This can be realized by chemical modification of the electrode surface or by integrating a drug delivery system in e.g. modified nanoporous platinum coatings. For loading the pores with active agents of various size, the pore diameter can be adjusted by the use of different templates like Pluronic® F127 and polystyrene latex beads (PLBs). To control the release behaviour of the active agents like rolipram or BDNF, which induced an enhanced SGN neuronal survival, the nanoporous platinum coating can be modified with several functionalized thiols.

Nanoporous platinum is deposited electrochemically on dense platinum surfaces using templates. Within the first method, Pluronic® F127 is dissolved in a Pt(IV) solution. During the pulsed potential electrochemical deposition of platinum, Pluronic® F127 is incorporated into the forming platinum coating and removed subsequently during calcination. These nanoporous platinum surfaces are modified with self assembled monolayers (SAMs) of different thiols to adjust the release behaviour of active agents. Within the second method, dense platinum surfaces are coated with PLBs. After template formation, the platinum is deposited in the voids of the PLB layer and the PLBs are removed. The nanoporous platinum coatings were characterized by SEM, sorption measurements, impedance spectroscopy, and cell culture investigations with NIH3T3 fibroblasts, spiral ganglion cells (SGNs) and bone-derived mesenchymal stem cells (BDMSCs). The thiol modification was determined via contact angle measurements as well as XPS.

Both methods lead to coatings containing nanopores. By using Pluronic® F127 pores with a size of about 10 nm are obtained, by using PLBs the average pore diameter corresponds to the size of the beads in the range of 50 to 500 nm (figure 1). Depending on the coating thickness, the specific surface area can be increased to 430 cm2·cm−2. Impedance measurements of the nanoporous platinum coatings show improved impedance behaviour in the lower frequency range. Cell culture investigations indicate a good cell compatibility and SGN neurite growth (figure 2). Release experiments with rolipram show an adjustable rolipram amount depending on the SAMs.

This work was supported by the DFG Cluster of Excellence EXC 1077/1 "Hearing4all".

Figure 1

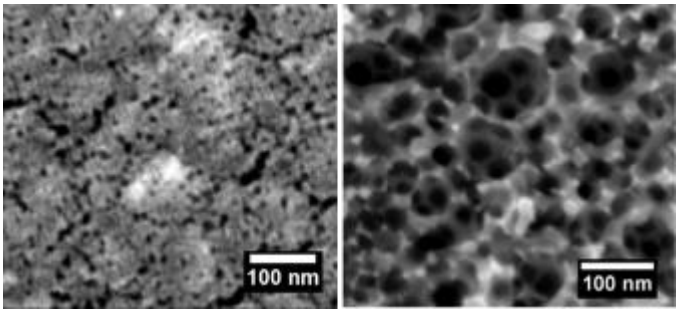
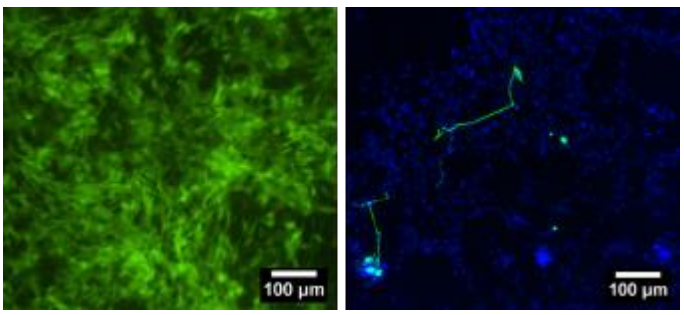


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P049

Synthesis of Fluorescent, Polysaccharide-modified Nanoporous Silica Nanoparticels for Bio Imaging Applications

*T. J. Pinkvos[1], I. Wille[1], C. Blume[2], P. Behrens[1]
[1]Leibniz Universität Hannover, Institut für Anorganische Chemie , Hannover, Deutschland
[2]Leibniz Universität Hannover, Institut für Technische Chemie , Hannover, Deutschland

Nanoporous silica materials offer a good biocompatibility, a large and permanent pore volume and versatility with regard to easily adjustable surface properties and are therefore often used within the field of drug delivery.[1,2] For tracking in bio-imaging applications, nanoporous silica nanoparticles (NPSNPs) can be equipped with fluorescent properties. To influence the fate of those particles with regard to their location and distribution in tissues or living systems, they can be modified with different polysaccharides. Our aim is to synthesize fluorescent NPSNPs with different polysaccharide coatings and observe their possible uptake and accumulation within vital cells.

In order to obtain such particles, we synthesized NPSNPs and modified them with fluorescin isothiocyanate (FITC) according to a synthesis by Lin *et al*.[3] In a second step the polysaccharides were attached to the surface.

As polysaccharides, we employed starch derivatives (provided by Rouqette GmbH), and polysialic acid (provided by the Institute of Technical Chemistry, Leibniz University of Hannover, Prof. Scheper).

Polysialic acid is involved in the development of neuronal tissue. The attachment happened according to a synthesis route, that has been worked out within our group.[1] To attach the starch derivatives we used a procedure developed by Bernardos *et al*.[4]

We were able to show the presence of the starch derivatives using the Tollens' test, while the Purpald® test showed the presence of the polysialic acid. Furthermore we were able to record a change concerning the pH-dependent zeta-potential due to the surface modification.

The fluorescence was observed under irradiation with UV light for the nanoparticles with attached FITC. Ongoing work focusses on the tracking and observation of the described nanoparticles in living cells.

This work was supported by the DFG Cluster of Excellence EXC 1077/1 "Hearing4all".

References

[1] S. Williams, A. Neumann, I. Bremer, Y. Su, G. Dräger, C. Kasper, P. Behrens, *J. Mater. Sci. Mater. Med.* **2015**, *26*, 125.
[2] Z. Li, J. C. Barnes, A. Bosoy, J. F. Stoddart, J. I. Zink, *Chem. Soc. Rev.* **2012**, *41*, 2590.
[3] Y.-S. Lin, C.-P. Tsai, H.-Y. Huang, C.-T. Kuo, Y. Hung, D.-M. Huang, Y.-C. Chen, C.-Y. Mou, *Chem. Mater.* **2005**, *17*, 4570–4573.
[4] A. Bernardos, L. Mondragón, E. Aznar, M. D. Marcos, R. Martínez-Máñez, F. Sancenón, J. Soto, J. M. Barat, E. Pérez-Payá, C. Guillem, et al., *ACS Nano* **2010**, *4*, 6353–6368.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P050

Platelet-Rich-Fibrin (PRF): Das *Low Speed Centrifugation Concept* (LSCC) als Grundlage für die Entwicklung von PRF-basierten festen und injizierbaren Blutkonzentraten.

*S. Ghanaati[1], J. Choukroun[2], S. Al-Maawi[1], R. Sader[1]
[1]Goethe Universität, Mund Kiefer und plastische Gesichtschirurgie , Frankfurt a. M., Deutschland
[2]Private Practice, Pain Therapy Center, Nice, France, , Nice, Frankreich

Hintergrund:

Platelet-Rich-Fibrin (PRF) wird durch die Zentrifugation des peripheren Blutes gewonnen. Mit Hilfe dieser Technik ist es möglich, flüssige und feste PRF-basierte Matrices ohne Zusatz von Antikoagulantien zu generieren.

Zielsetzung:

Ziel der Untersuchungsreihe war es, den Einfluss der Zentrifugalkraft in drei Bereichen (hoch, mittel und niedrig) auf die Zusammensetzung der festen und flüssigen PRF-Matrices zu untersuchen.

Material und Methoden:

Nach Herstellung von festem und flüssigem PRF in drei Zentrifugationsprotokollen, wurden die Blutkonzentrate in vitro kultiviert und die Überstände für die Proteinquantifikation gesammelt. Die Zell- und Proteinverteilung in den flüssigen und festen PRF-Matrices wurde mittels ELISA, Durchflusszytometrie und Histologie bestimmt.

Ergebnisse:

In der flüssigen PRF-Matrix mit niedriger Zentrifugalkraft zeigte sich eine signifikant höhere Zellzahl (Thrombozyten und Leukozyten) als in den PRF-Matrices mit mittlerer und hoher Zentrifugalkraft. Im festen PRF mit niedrigerer Zentrifugalkraft zeigte sich eine gleichmäßige Verteilung der Zellen, wohingegen in PRF-Matrices mit mittlerer und höherer Zentrifugalkraft die Zellen nur in unteren Bereichen der PRF-Clots lokalisiert waren. Die Wachstumsfaktorfreisetzung (VEGF, TGF ß-1 und EGF) in den festen und flüssigen PRF-Matrices war in den Protokollen mit niedrigen Zentrifugalkraft signifikant höher als in PRF-Matrices, welche mit mittlerer und höherer Zentrifugalkraft hergestellt worden sind.

Schlussfolgerung:

Die Reduktion der Zentrifugalkraft ermöglicht die Herstellung von bioaktiveren Blutkonzentraten, welche signifikant mehr Zellen beinhalten und höhere Konzentrationen an Wachstumsfaktoren freisetzen. Wir postulieren das Low Speed Centrifugation Concept (LSCC) als eine Möglichkeit zur Herstellung patientenspezifischer Blutkonzentrate, welche als ein autologes "drug delivery system" dienen und Biomaterialien bei Ihrem Beitrag zur Wundheilung und Geweberegeneration unterstützen könnten.

Literatur

[1] S. Ghanaati et. al. J. Oral Implantol. 40 (2014) 679–689.
[2] K. El Bagdadi et al. Eur. J. Trauma Emerg. Surg. (2017).
[3] J. Choukroun, S. Ghanaati. Eur. J. Trauma Emerg. Surg. (2017).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P051

Hierarchical scaffolds with meso and macro porosity consisting of bioglass, gelatin and chitosan

*F. Schmidt[1], L. M. Henning[1], S. Zavareh[1], F. Zemke[1], P. Kamm[2], J. Banhart[2], A. Gurlo[1]
[1]Technische Universität Berlin, FG Keramische Werkstoffe , Berlin, Deutschland
[2]Technische Universität Berlin, FG Struktur und Eigenschaften von Materialien , Berlin, Deutschland

Bone consists of a natural composite of organic (collagen) and inorganic (bone mineral ydroxyapatite) phases with a highly porous structure. Researchers try to mimic the hierarchical, porous and/or material structure of bone to enable improved regeneration in case of trauma or illness. Artificial bone replacement materials should fulfill several requirements, such as suitable mechanical properties, and good biocompatibility, ideally even bioactivity. Furthermore the geometry should mimic that of natural bone, e.g. high porosity coupled with high surface area in order to encourage cell adhesion, proliferation and the transport of nutrients into the newly forming tissue.

We have applied freeze casting as a simple and economic method to produce macroporous scaffolds with 75 % porosity and good mechanical properties with up to 75 vol.% bioactive glass content. Gelatin and chitosan might possess thrapeutical properties and therefore have been used as binder and dispersant, respectively.

The processing method applied allows for great control over macroporosity, solid content, pore size, and pore structure by varying slurry composition and processing routes. It is further possible to implant a drug releasing system when applying mesoporous bioactive glasses. The resulting scaffolds show mesoporosity for improved cell adhesion and possible drug release as well as macroporosity for cell ingrowth and nutrient transport into the scaffold.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P052

Multinucleated giant cells into the implant bed of bone substitutes are foreign body giant cells – consequences for bone tissue regeneration

*M. Barbeck[1,2], R. E. Unger[3], S. Stojanovic[4], S. Najman[4], F. Witte[2]
[1]botiss biomaterials GmbH, Research & Development , Zossen, Deutschland
[2]Charité - Universitätsmedizin Berlin, Julius Wolff Institute and Center for Musculoskeletal Surgery, Berlin-Brandenburg Center for Regenerative Therapies , Berlin, Deutschland
[3]University Medical Center of the Johannes Gutenberg University, Mainz, Institute of Pathology , Mainz, Deutschland
[4]University of Nis, Faculty of Medicine , Nis, Serbien

Introduction

Biomaterial-associated multinucleated giant cells (BMGCs) have been found within the implantation beds of most bone substitutes irrespective of their origin, i.e., allogeneic, xenogeneic or alloplastic materials. However, their exact differentiation and their involvement in healing events still remain mostly unclear. Various findings suggest that these cells belong to the cell line of the foreign body giant cells (FBGCs), which are of "inflammatory origin". Interestingly, they may provide a phenotypic heterogeneity equivalent to that of macrophages, whose anti-inflammatory activation profile has been proposed to promote the process of biomaterial-mediated tissue regeneration [1]. Thus, further knowledge is essential to evaluate the role of BMGCs in the bone regeneration processes as this is required to ensure their successful clinical application.

Methods

Two different studies were conducted to analyze both the differentiation of the BMGCs and their heterogeneity. In a first study tissue samples from a clinical study were used to analyze the origin of BMGCs in the implant beds of a synthetic and a xenogeneic bone substitute. Two antibodies against integrin molecules specific for osteoclasts (β-3 integrin) or FBGCs (β-2 integrin) were used to distinguish both giant cell types. In a second study an established subcutaneous implantation model in 24 female Wistar rats to implant silk fibroin (SF) was used. Specialized (immuno-) histochemical staining methods and histomorphometrical techniques, were applied to analyze the heterogeneity of BMGCs [2].

Results

The results of the first study indicate that the BMGCs induced by both kinds of bone substitutes are FBGCs, as they express only β-2 integrin in contrast to the osteoclasts outside of the immediate implantation areas, which only demonstrate β-3 integrin expression (Fig. 1). Furthermore, these cells express both pro- and anti-inflammatory molecules to the same extent within the implantation beds of silk fibroin scaffolds 15 days after implantation.

Conclusion

These data give new insight into the tissue reaction to biomaterials. Based on this new knowledge further research concerning the proteomic profile of the FBGCs especially with respect to the different physicochemical properties of biomaterials is necessary.

Figure 1

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P053

Functionalization of polymer fibers with extracellular matrix components

*I. Wille[1], P. Behrens[1], J. Schulze[2], A. Warnecke[2]
[1]Leibniz Universität Hannover, Institut für Anorganische Chemie , Hannover, Deutschland
[2]Hannover Medical School, Department of Otorhinolaryngology , Hannover, Deutschland

The application of polymer fibers consisting of poly-ε-caprolactone (PCL) and polyglycolide (PGA) has already been widely studied in the field of biomedical sciences. Both polymers have been used for example as basic material for drug delivery systems or 3D-scaffolds in tissue engineering. They allow an efficient modification of their physical, chemical and mechanical properties by copolymerization or blending with many other polymers as well as by surface functionalization. Moreover they have shown a good biocompatibility and are biodegradable.[1] The aim of this work is to modify the surface of fibers made from PCL/PGA copolymers in order to test their applicability as components in scaffolds for tissue regeneration. For this purpose, the biodegradable fibers were coated with heparan sulfate as a representative of extracellular matrix components; heparan sulfate is also an anticoagulant and could enhance wound healing after implantation.[2] It is well-known in clinical applications and is, for example, used in the eyedrops Cacicol® to regenerate and heal the cornea.

This work was supported by the Cluster of Excellence Hearing4all.

In this work the polymer fibers are functionalized with aminogroups via aminolysis using ethylenediamine.[3] Afterwards heparan sulfate is linked covalently to the functionalized fibers using coupling and stabilizing agents.[4] Toluidine blue was used in a colorimetric assay to validate the presence of heparan sulfate on the fibers.[5] Furthermore, cell culture investigations were done.

Both substances could be attached to the surface of the polymerfibers. A release of laminin was observed. Cell culture investigation showed a good cytocompatibility.

As there are promising results, more in vitro and in vivo studies will follow to test the applicability of the synthesized material.

References

[1] Yoo, H. S., Kim, T. G., Park, T. G.; *Adv. Drug Deliv. Rev.*; **2009**; 61; 12; 1033–1042.
[2] Rabenstein, D. L.; *Nat. Prod. Rep.*; **2002**; 19; 1 4; 312–331.
[3] Huang, Y.-C., Huang, C.-C., Huang, Y.-Y. & Chen.; *J. Biomed. Mater. Res.;* **2007***;* 82A*;* 842–851.
[4] Zhu, Y., Leong, M. F., Ong, W. F., Chan-Park, M. B., Chian, K. S.; *Biomater.*; **2007**; 28; 861–868.
[5] Pieper, J. S., Van Wachem, P. B., Van Luyn, M. J. a, Brouwer, L. a., Hafmans, T., Veerkamp, J. H., Van Kuppevelt, T. H.; *Biomater.*; **2000**; 21; 1689–1699.
[6] Macintosh, F. C.; *Biochem. J.*; **1941**; 35; 776–82.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P054

Microstructured glass for biomedical applications - Subsurface laser scribed channels and cavities

*U. Brokmann[1], R. Schade[2], K. Liefeith[2], E. Rädlein[1]
[1]TU Ilmenau, Group of Inorganic-nonmetallic materials , Ilmenau, Deutschland
[2]Inst. für Bioprozess- und Analysentechnik e.V., Department of biomaterials , Heiligenstadt, Deutschland

Photosensitive glasses are well suitable materials for tissue engineering applications due to their high chemical and thermal stability and their biocompatibility. The standard procedure for the fabrication of several scaffold types is the 3 step photoform process, consisting of an exposure step for the photochemical modification of a glass, a thermal treatment step for the crystallization of a lithium metasilicate phase and an etching step in diluted hydrofluoric acid for the generation of cavities in the exposed and crystallized areas. Cavities at the micro and nano meter scale have been inscribed into the material using focused laser radiation. It was found, that single photon excitation as well as multi photon excitation can be used to modify different glass components for the fabrication of complex three dimensional burried geometrical structures. Material properties like size and distribution of lithium metasilicate crystals affect process values like aspect ratios of geometrical structures or the surface roughness in etched grooves. Micro mechanical, fluidic or optical elements in biological applications require different strategies for the optimization of these parameters.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P055

Effective strategies for controlling and dealing with transmission pathways of pathogens in air traffic using biomaterials surfaces

*B. Zhao[1], C. Dewald[1], M. Hennig[1], J. Bossert[1], K. D. Jandt[1]
[1]Friedrich Schiller University Jena, Chair of Materials Science, Otto Schott Institute of Materials Research , Jena, Deutschland

Aircrafts transport billions of civilians over vast distances around the world in a short time, which undoubtedly benefits tourism and commerce under the increasing globalization trends. However, air travels may also contribute significantly to the rapid spread of infectious diseases that were geographically confined in the past, creating international epidemics with great health and socio-economic impact. Possible influences on the spreading of pathogens via air travel are climate, infrastructure and indirect transmission pathways, e.g. materials surfaces.

The HyFly project aims to understand and interrupt the infection routes in air traffic. Therefore, an analysis of risk factors for infection transmission across sectors needs to be performed for aircrafts and airports. This will also be assessed based on climate properties. Furthermore, the project aims to reduce infection transmission by structural constructive solutions. Different subprojects focus on solutions to improve cleaning strategies, materials surfaces and diagnostics applicable in air traffic.

A literature review is carried out to investigate the contamination, survival and transmission of pathogens via the surfaces of interior construction materials in aircrafts, suggesting that the door handles, armrests, tray tables or lavatory components could be a potential risk for infection. Therefore, one of the subprojects will develop an effective strategy for infection control by optimizing the surface properties (compositions, crystallinity, topography, roughness, wettability, etc.) of materials within aircrafts.

Acknowledgements

This work was supported by the InfectControl2020 HYFLY project "Effective strategies for the controlling and dealing with transmission pathways of pathogens in air traffic" which is funded by the Federal Ministry of Education and Research (BMBF), Germany.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P056

Development of Tumor-like Microcapsules with tunable biodegradability and mechanical properties to study single cell intravasation and metastasis *in vitro*.

L. Höne[1], A. Schwab[1], B. Fabry[1], A. R. Boccaccini[1], *A. Leal-Egana[1]
[1]Universität Erlangen-Nürnberg, , Erlangen, Deutschland

Cancer is one of the main causes of mortality worldwide. With the purpose to understand cancer progression *in vitro*, we are currently developing biocompatible scaffolds resembling the neoplastic niche.

Recently, we improved traditional Tumor-like microcapsules(1) to analyze the role that three-dimensional mechanical stimulation plays on intravasation and further metastasis.

In our work, we encapsulate a very low amount of MCF10A, BT-549 or MCF7 cells in Tumor-like microcapsules (approximately 10 cells per bead). These scaffolds were made of alginate/gelatin (1:1 blend). This composition was chosen with the purpose to submit entrapped cells to mechanic and metabolic stress. In addition, this experimental design allowed us to observe: migration and proliferation within the scaffolds at single-cell scale, formation of entrapped aggregates from single cells, and detection of single cells/aggregates undergoing to intravasation.

Our results have shown that all cell types were able to migrate and proliferate within these 3D scaffolds. However, just a few clones of MCF7 cells were able to abandon the tumor-like microcapsules and to colonize 2D microenvironments. Cells colonizing 2D surfaces, exhibited biophysical differences to pre-entrapped cells. According to our results, selection of cells able to intravasate was driven by the mechanical stress suffered by entrapped cells.

It is interesting to note that, after working with different migrating cancer cells on patterned hydrogels, we recently reported the existence of sub-clones expressing specific biophysical biomarkers(2). These markers made possible the differentiation between metastatic and non-pathological cells(2). Thus, in this work we hypothesize that a similar trend could be studied in cells entrapped within 3D neoplastic-like niches, considering the possibility that just a few clones, found within an entire population, could have the capability to proliferate and to migrate out from the Tumor-like microcapsules.

References

[1] Leal-Egana et al., (2012). Tuning liver stiffness against tumours: an in vitro study using entrapped cells in tumour-like microcapsules. Journal of the Mechanical Behavior of Biomedical Materials 9: 113-121.

[2] Leal-Egana et al., (2017). The size-speed-force relationship governs migratory cell response to tumorigenic factors. Molecular Biology of the Cell. In the press. DOI: 10.1091/mbc.E16-10-0694.

Figure 1

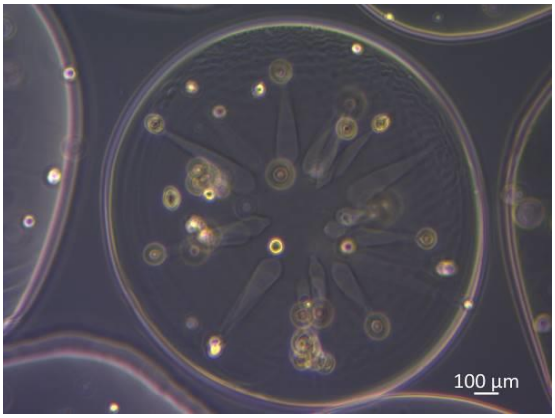
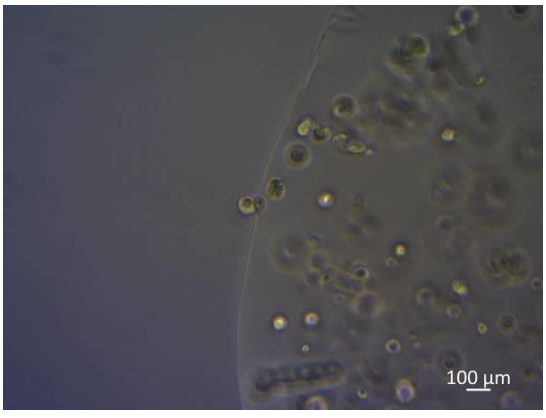


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P057

HistoGap – morphometric-software for 2D image data with Application focus on quantitative evaluation of Osseointegration from Implants

*C. Müller[1], D. Linde[1], S. Kottke[1], K. Otto[1], S. Voigt[1], G. Schneider[1]
[1]Universitätsklinikum Jena, HNO-Forschungslabore, AG Biomateriallabor , Jena, Deutschland

Aim

A significant criterion for the evaluation of the biocompatibility of implants in bone is their Osseointegration. The bone-implant-distance (BID) gives quantitative statements about that.
The BID of cylinder-shaped implants embedded in bone was determined by measuring hard-tissue-histology slides with the self-developed morphometric software "HistoGap". A precise measurement of the distance is only possible if the cut runs exactly by the cylinder axis, which is practically not feasible.
To compensate the resulting divergence, an algorithm was developed, implemented and tested.

Method

In vivo Tests according to the "Jenaer Schädelmodell" (Jena skull model) were perform in New Zealand White Rabbits. After explantation of the implants with surrounding tissue, hard-tissue-histology slides were prepared [1].
To evaluate these 2D image data quantitatively, implant and BID were measured in detail using HistoGap.
The measuring values were automatically corrected by the software and for further evaluation exported together with data of the experiments conditions.

Results

Supported by the ergonomic user interface of HistoGap, the hard tissue histology image data were evaluated precisely and efficiently.
The proportion of the implant surface with direct bone contact (Bone-Implant-Contact, BIC), which is considered as "gold standard" for the assessment of Osseointegration, can be directly derived from the measured BID values.

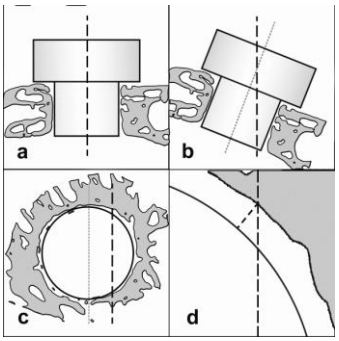
Conclusion

The HistoGap morphometric software allows fast and uncomplicated measurement of approximately parallel arranged structures in 2D images (e.g., gaps, cracks, layers).
With HistoGap, the relevant experimental conditions for the subsequent statistical evaluation are stored in structured form, which allows detailed analyzes of complex experimental setups.
Particularly when evaluating the Osseointegration of cylindrical implants, the correction of the geometrically caused systematic measurement errors brings significant advantages:
Quantitative results with a higher statistical significance can be derived from a limited number of experimental data.
In addition, sections with a strong deviation from the optimal orientation, which could not yet be evaluated, can be included in the quantitative evaluation. Thus, the experimental data base is significantly broadened.

References

[1] Schneider G (2010) J Mater Sci Mater Med 21(10):2853

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P058

The influence of cobalt ions on collagen assembly and cell fate

*E. McCarthy[1], H. Floyd[1], L. M. Grover[1]
[1]Univeristy of Birmingham, Chemical Engineering , Birmingham, Vereinigtes Königreich

Metal-on-metal (MOM)-based implants are the gold standard for hip joint replacements due to their robustness. However, MOM implants wear over time to produce small metal particulates and metallic ions that infiltrate the surrounding tissue, leading to local inflammation, necrosis and ultimately implant failure. The extracellular matrix, which comprises mainly of collagen type I, is a key part structural component of tissues and capable of directly influencing cell function. Understanding the effect of wear debris on the matrix, rather than just direct effects on cells, may provide a new insight into the mechanism of implant failure and implant-tissue bonding.

To assess the effect of cobalt II ions on the gelation of collagen type I and corresponding influence on the fate of MC3T3-E1 cells.

Cultrex® rat collagen I hydrogels with varying concentrations of cobalt II ions were created. The micron structure was imaged using an Innova NanoScope Atomic Force Microscope, using tips with a frequency of 20 kHz and a force constant of 0.9N/m. Bulk properties were determined via an ARG-G2 Rheometer, with a frequency sweep from 0.01-10Hz performed at a strain of 0.1. Water retention was determined by measuring the weight loss in the various hydrogels over a series of 5 hours. MC3T3 cells were seeded onto the hydrogels and imaged via a FV10-ASW confocal microscope using Calcein AM and Propidium Iodide dyes to determine cell viability. An AlamarBlue® assay was also undertaken from the same sample set up and read 4 hours later at an absorption emission of 570nm.

On the micron scale, cobalt ions increase the heterogeneity of the matrix suggesting a cross-linking interaction between the collagen fibrils and the cobalt ions. This was confirmed by the observation of decreased water retention as Cobalt ion concentration increased. Rheology results showed that the addition of Cobalt ions significantly reduced the stiffness of the bulk structure. The Alamar Blue assay and the live/dead confocal imaging showed that increasing Cobalt ion concentration reduced cell viability and proliferation.

Cobalt ions interact with collagen fibrils altering the localised and bulk structure of the collagen matrix, causing a decrease in cell proliferation and cell viability. Therefore, the mechanism of MOM implant failures could also include "chemical remodelling" and weakening of the extracellular matrix.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P059

Measuring thermal degradation during melt electrospinning writing

*C. Böhm[1], A. Hrynevich[1], G. Hochleitner[1], J. Groll[1], J. Teßmar[1], P. D. Dalton[1]
[1]University Hospital Würzburg, Department for Functional Materials in Medicine and Dentistry , Würzburg, Deutschland

Introduction: Solution electrospun scaffolds is widely used to investigate cell behavior[1]. One disadvantage of the process is that organic solvents (often cytotoxic) are often required. A non-solvent alternative is called "Melt Electrospinning Writing" (MEW) that uses polymer melts instead of solutions to produce 3D scaffolds with improved precision of fiber placement. One limitation with MEW is the requirement of low flow rates while elevated temperatures can result in degradation of the polymer. This investigation uses specific printing conditions to allow 1) monitoring of degradation and 2) predicting of the degradation when new polymers are processed via MEW.

Materials and methods: Polymers (PCL and PLGA) are heated in a glass syringe and loaded into a custom-built MEW printer. The syringe is purged with nitrogen gas for 15 min to prevent oxidation or hydrolysis during heating. The melt is driven out to the spinneret by nitrogen pressure, and a high voltage applied to the spinneret. An electrified molten jet is direct-written over a collector, first stabilized and then as arrays on a microscope cover slip, that were divided into 9 blocks with 4 arrays each.

Results: MEW could be performed in two modes – linear or non-linear, depending on the collector speed[2]. When the speed of the collector matches or is above the speed of the jet, straight lines result. For collector speeds slower than the jet, non-linear fibers with patterns are generated. The speed where the collector and jet are equal is referred to as the "Critical Translational Speed" (CTS). Monitoring the shape of the fiber deposited around the CTS allows observation of thermal degradation of the polymer over time.

Conclusion: Observing shifts in the CTS allows investigation of thermal degradation of molten polymers. This method of monitoring the CTS and resulting deposition patterns enables early determination of thermal degradation under actual printing conditions. Future research is required to determine whether it is the generation of charged species and/or viscosity reduction that results in changes in jet behavior with time.

References

[1] T. D. Brown, C. Vaquette, D. W. Hutmacher, P. D. Dalton, in *Polymeric Materials: Structure and Function, Vol. 1* (Eds.: S. Dumitriu, V. Popa), **2013**, pp. 539–592.

[2] G. Hochleitner, A. Youssef, A. Hrynevich, N. Haigh Jodie, T. Jungst, J. Groll, D. Dalton Paul, *BioNanoMaterials* **2016**, 10.1515/bnm-2015-0022.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P060

Synthesis, coating and *in vitro* study of antimicrobial copolymers for dental implants

*S. Stelljes[1], M. Waßmann[1], A. Winkel[2], M. Stiesch[2], H. Menzel[1]
[1]TU Braunschweig, Institute of Technical Chemistry , Braunschweig, Deutschland
[2]MH Hannover, Clinic of Dental Prosthetics and Biomedical Material Science , Hannover, Deutschland

Background

Dental implants are an important tool in today"s dental medicine. However, there are still problems to overcome. The mouth is ideal for bacteria to grow because of humidity, temperature and food residues. This may lead to biofilm formation and inflammatory responses like periimplantitis, which is the case for 8-10% of all implants[1]. Using copolymer coatings to prevent biofilm formation, but also enable human cells to grow might be a promising approach. Such copolymers are built from momomers carrying antimicrobial moieties like quaternary ammonium[2] or guanidine groups[3] and phosphonate groups to attach to the implant[4].

Methods and Results

Two copolymers containing quaternary ammonium groups (P(DMAEMA-co-DMMEP) and (P(VI-co-DMMEP) and one copolymer containing guanidine groups (PEDBEG-GMA-co-DMMEP) were prepared via free radical polymerization. All three polymers were coated onto titanium disks using spin coating and then heated to ensure covalent attachment to the disks. Layer thickness measurement via ellipsometry and water contact angle measurements were carried out to characterize the coatings.

Cytotoxicity analysis for all copolymers in solution show no cell damaging properties in LDH- and CellTiter-Blue assays. Human gingiva fibroblasts (HGFib), *S.aureus* and *S.oralis* were cultivated on coated disks to analyze cell adhesion and antimicrobial effect. All coatings show a strong antimicrobial effect against both germs. Cell adhesion is poor for DMAEMA and VI-Copolymers while PEDBEG-GMA-co-DMMEP shows very good adhesion of HGFib compared to titanium. Coated disks were sterilized with gamma radiation and steam pressure sterilization. This has only little effect on antimicrobial capabilities and layer thickness for DMAEMA- and VI-containing copolymers. For the PEDBEG-copolymer antimicrobial effect and layer thickness are affected by gamma irradiation, but both remain nearly unaffected upon pressure sterilization.

In conclusion, all three copolymer coatings presented in this study show good potential to be used as coatings on dental implants and will be used for *in vivo* experiments in the near future.

References

[1] Pjetursson, B.E., Thoma, D. et al; *Clin Oral Implan Res*; 23; **2012**; 22-38.
[2] Tiller, J.C., Kilbanov, A.M. et al; *PNAS*; 98(11); **2001**; 5981-5985.
[3] Ikeda, T., Hirayama, H., Watanabe, M., et al; *Antimicrob. Agents Chemother.*; 30(1); **1986**; 132-136.
[4] Callies, T., Bartsch, I., Menzel, H. et al; *Mater Sci Eng C*; 61; **2016**; 712-719.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P061

Modified Bacterial Nanocellulose as biodegradable carrier system for antibiosis in dentistry

*U. Beekmann[1], P. Weyell[1], N. Reinhöfer[2], D. Kralisch[1]
[1]Friedrich-Schiller-University, Department of Pharmaceutical Technology , Jena, Deutschland
[2]University Hospital, Center of Stomatology , Jena, Deutschland

Question

Bacterial nanocellulose has shown a high potential as innovative drug delivery system [1, 2]. We recently investigated the potential of drug-loaded hydrated and freeze-dried BNC in dental therapies such as dental extraction or mucosal transplantation, *e.g*., for the palate or the extraction alveole coverage. Both applications would benefit on the one hand from a material, which degrades under physiological conditions and on the other hand from an antibiotic environment.

Methods

Consequently, periodate-oxidation of BNC was performed to facilitate modified oral degradation behaviour. To ensure biocompatibility of oxidized BNC, an *in-vitro* toxicity test (MTT assay) was implemented. In addition, native and oxidized BNC loaded with doxycycline was tested for prophylaxis against infection. To prove antibiotic efficiency against pathogen oral bacteria such as *Staphylococcus aureus, Aggregatibacter actionmycetemcomitans, Streptococcus mutans*, agar diffusion tests of BNC samples loaded with doxycycline *completed the methods.*

Results

Through periodate-oxidation, oral degradation was realised and controllable. MTT assay proved preserved biocompatibility of the chemically modified biomaterial. Comparative release studies of the drug from native and oxidized BNC have shown that the release behaviour is only marginally affected by the oxidation. Agar diffusion test confirmed antibiotic efficiency of the biodegradable drug delivery system.

Conclusions

In summary, BNC has proven once more as a highly adaptable carrier for the design of tailor-made drug delivery systems.

References

[1] S. Moritz, et al.; *Int. J. Pharm*., 471 (2014), 45-55
[2] Y. Alkhatib, et al.; *Eur. J. Pharm. Biopharm*., 112 (2017), 164-176

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P062

Development of a new generation bone substitute material tested in a rat tibia defect model

*C. Ganz[1], M. Dau[2], F. Zaage[1], W. Xu[1], B. Frerich[2], T. Gerber[1]
[1]Universität Rostock, Institut für Physik , Rostock, Deutschland
[2]Universitätsmedizin Rostock, Klinik und Poliklinik für Mund-, Kiefer- und Plastische Gesichtschirurgie , Rostock, Deutschland

Bone replacement materials are used in many medical fields, for example in dentistry, surgery and orthopedics as an alternative to autologous bone, which is still considered as a gold standard. Bone sustitutes include autografts, xenografts, allografts or synthetically manufactured materials. NanoBone® (Artoss GmbH Rostock, Germany) is an established synthetic bone grafting material that is degraded by osteoclasts. Furthermore, the composition of the material changes after implantation. SiO2 as a component is replaced by an organic matrix (matrix changes). [1]. However, for an aesthetic application in dentistry, it is utile to use a material that is slowly resorbed.

Therefore, the aim of this work was to develop a new generation of bone grafting material and to test it in an appropriate *in vitro* and *in vivo* model to analyze cell reactions, bone formation and degradation of biomaterial via remodeling processes. The new NanoBone® is a nanocrystalline hydroxyapatite (HA) embedded in a highly porous matrix of silica (SiO2) gel, based on Sodium Waterglass (Sodium Silicate) at the ratio of 96:4 (wt%) HA:SiO2.

In vitro cytotoxicity studies were performed on mouse fibroblasts (L929) via an indirect XTT test. No toxic reactions of the cells to the material were observed. *In vivo* investigations were performed on an established rat tibia model [1]. Male Wistar rats (body weight 400-500g) were used for the experiments. After a 2cm skin incision, a 3.5mm diameter round cortical defect was created using a dental drill under constant irrigation with saline. These defects were filled with the biomaterial. The animals were sacrificed at 12, 21 and 63 days respectively (n=5 for each time point). Decalcified thin slices were prepared, followed by a histological and histomorphometric evaluation of the samples. After 12 days 18.6 ± 2.1% of newly formed bone could be detected. After 63 days, the amount of bone increased to 28.9 ± 0.4%. The amount of bone substitute material did not change over the trial period. This suggests that the tested biomaterial was not remodeled through osteoclasts. The results show that the reduction in the concentration of SiO2 has a significant influence on degradation.

References

[1] Ganz C,Xu W, Holzhüter G, Götz W, Vollmar B, Gerber Th: Comparison of bone substitutes in a tibia defect model in Wistar-rats. *Key Engineering Materials Vols. 493-494 (2012) pp 732-738*

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P063

Heat Treatment Simulation of TiZr Alloy with Application of Phase Field Method

*A. Fotovati[1], S. Schmauder[1]
[1]Universität Stuttgart , IMWF , Stuttgart, Deutschland

TiZr alloy is a great candidate for small diameter implants, even in high loading situations. It is biocompatible, has a good corrosion resistance with a osseointegration and has a higher strength of 40% with respect to commercially pure titanium (cpTi). TiZr has a perfect solid solution at 47.8 wt.% Zr according to coherent scattering map. According to these facts we used the Phase Field Method (PFM) for simulating the microstructure of TiZr where at different temperatures the α phase (800K), α+β phases (923K) and only β phase (1200K) are stable at 47.8 wt.% Zr. A single crystal with dimension of 20x20 nm is used as simulation box. Both spacial and temporal evolution of the microstructure are solved according to FEM with help of Generic Phase Field Library of MOOSE software. Free energy equation of each specimen is extracted from the CALPHAD database with the help of Thermocalc® Software.

As we expected after about 1 hour at 800K a random distribution of α nuclei are growing in the β matrix and completely covering the box of simulation (Fig.1). At 923K, α nuclei are growing till they cover 87% of the matrix (Fig.2). Finally, at 1200K, α nuclei very fast dissolve in the β matrix. Results show that PFM not only can predict the microstructure as we expected from the phase diagram of TiZr, but also show the morphology of each phase that can be used as a start point for mechanical simulation. Moreover, without expensive laboratory tests, it is possible to predict the evolution of the microstructure for different compositions and temperatures.

Figure 1

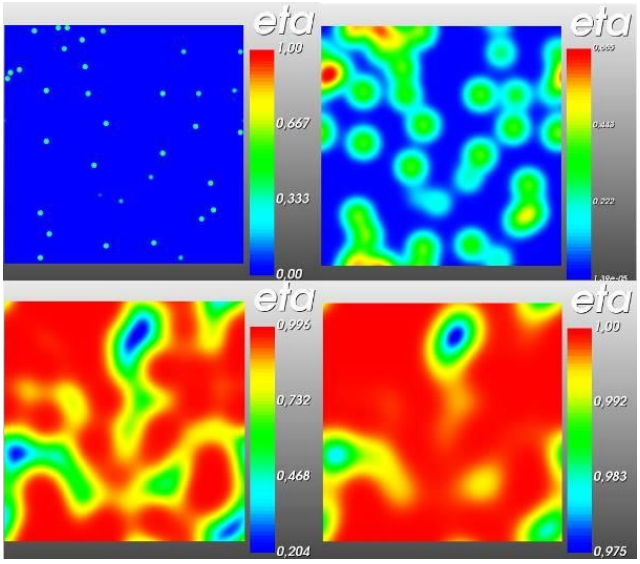
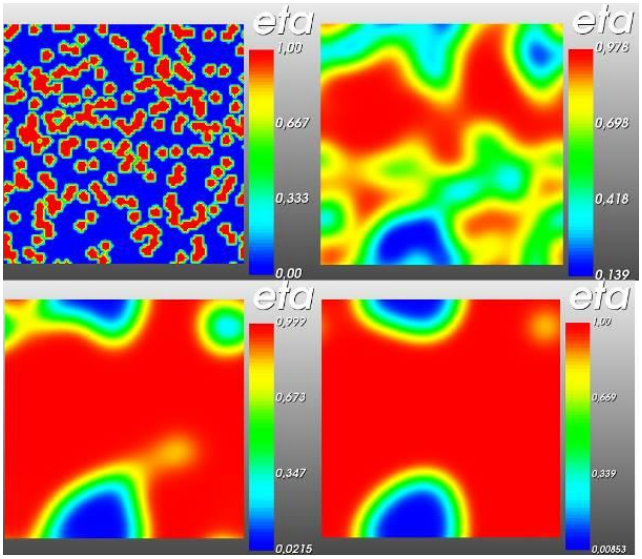


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P064

Influence of a mouthwash containing *Casearia sylvestris* on color alteration and surface roughness of tooth

*L. Oliveira Lima Bohner[1], A. P. Terossi de Godoi[2], A. Blásida Concepción Elizaur Benitez Catirse[3]
[1]University of São Paulo, , Sao Paulo, Brasilien
[2]Uniararas, , Araras, Brasilien
[3]School of Dentistry of Ribeirão Preto, University of São Paulo, , Ribeirão Preto, Brasilien

Introduction: In order to overcome Clorhexidine 0,12% adverse effects, a mouthwash contaning *Casearis sylvestris* was developed. **Objectives:** The purpose of this study was to evaluate the influence of a mouthwash containing *Casearia sylvestris* on color alteration and surface roughness of teeth previously submitted to bleaching or in contact with acid drinks. **Material and Methods:** Thirty cylindrical bovine teeth samples were divided in 3 groups, according to the solution evaluated: A) distilled water, B) Chlorhexidine 0,12% and C) a mouthwash containing *Casearia sylvestris*. Each group was divided in subgroups, according to the categories (n=10): 1) No treatment (control group), 2) bleached teeth and 3) teeth immersed in an acid drink (pH 2,4). The bleaching procedure was performed with carbamide peroxide 16% during 6 hours by 8 days, totalizing 48 hours of immersion. Whereas the acid drink immersion was performed once in a day during 2 minutes each immersion, for 6 days. Afterward, the samples were immersed into a bowl containing the respective solutions by 2 minutes, three times per day, during 7 days. The color and surface roughness were evaluated before the first immersion on the solutions and after 7 days of immersion. In addition, a qualitative analysis was performed by scanning electron microscopy. **Results:** Data were statistically analyzed by two-way analysis of variance (ANOVA) and Tukey`s test. Teeth submitted to bleaching or immersed in acid drinks presented a higher color alteration in comparison to the control group. Clorhexidine 0,12% (B1-0.21±0.08, B2-0.84±0.08, B3-0.69±0.08) and the mouthwash containing *Casearia sylvestris* (C1-0.53±0.08,C2-0.71±0.08, C3-0.74±0.08) did not provide a statistically higher color alteration in comparison to the distilled water (A1-0.0±0.08; A2-0.72±0.08, A3-0.80±0.08). The surface roughness was not altered by the solutions and treatment provided. **Conclusion:** The mouthwash containing *Casearis sylvestris* can be safely used on teeth previously submitted to bleaching or immersed in acid drinks during the period evaluated in this study.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P065

The effect of thranekron and autograft bone on bone defect healing in rabbit model

*A. Bighamsadegh[1], S. Sharifi[1], A. Oryan[2], Y. Alavi[1]
[1]Shahrekord University, Veterinary Surgery , Shahrekord, Iran
[2]Shiraz University, Pathobiology , Shiraz, Iran

Introduction: Nowadays, bone grafting is used in both human and veterinary orthopedics to stimulate fracture healing, accelerate joint union, and to restore bone defects. In such procedures, orthopedic surgeons are searching to favorable substitute for autograft bone. The present study seeks to investigate the use of this extract in bone defect healing process.

Methods: 20 native rabbits were divided into 4 groups of five, and bone fragments were removed from their radius bones. In the first group, Theranekron 1mg/kg was injected into bone defect. Normal saline was injected into the same site of the five rabbits in the second group (saline treated controls). As for the third group (the untreated controls), nothing was injected. In the fourth group (autograft group), the removed bone was put in place and muscle and skin were sutured. Post op lateral x-rays were prepared on days 14, 28, 42, and 56. Bone biopsy for histopathological study was performed after 8 weeks. Radiographs and samples were analyzed statistically in terms of union, osteogenesis activity, and bone Remodeling.

Results: Radiological evaluation showed that autograft group was significantly superior to both untreated and saline treated controls (p=0.02, p=0.04, respectively). Also, Theranekron group proved better than untreated controls (p=0.04). Histopathological evaluation showed terabecular bone and bone marrow formations in Theranekron and also autograft groups. In the untreated and saline treated controls, most part of defect was filled with fibrous tissue.

Conclusions: Theranekron treated group showed almost similarly to autograft counterpart, but better than normal saline and untreated controls.

Figure 1

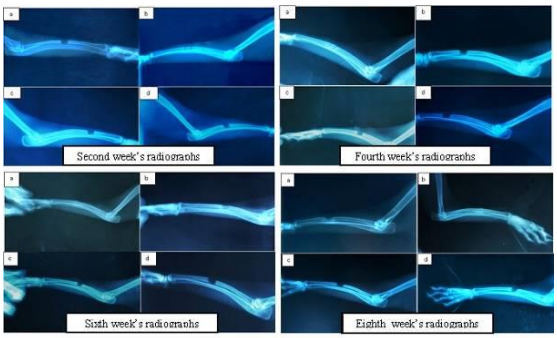
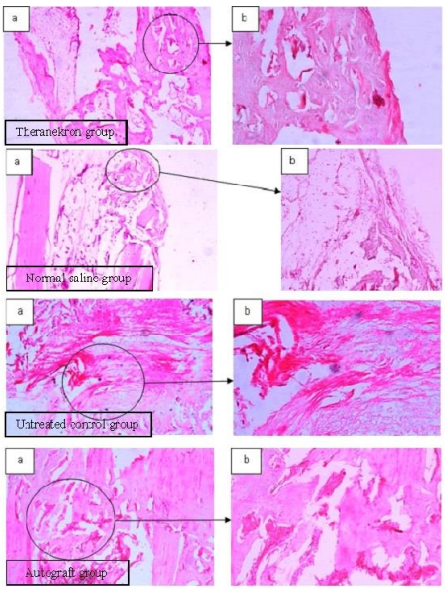


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P066

Effects of allogenic and xenogenic cartilage graft on bone defect healing in rabbit model

*A. Bighamsadegh[1], S. Sharifi[1], A. Oryan[2], I. Karimi[1], F. Hoseini[1], M. Maleki[1]
[1]Shahrekord University, Veterinary Surgery , Shahrekord, Iran
[2]Shiraz University, Pathobiology , Shiraz, Iran

Introduction

Orthopedic surgeons are trying to find best substitutes for bone grafting in human and veterinary medicine. Bone autografts are still as a golden standard in comparison with other bone grafts. Diced ear cartilage from dog and rabbit have been used on bone healing previously, in the present study effects of rabbit and bovine rib cartilages will evaluated in bone healing of rabbit bone defect model.

Method

In this study, 20 adult rabbits weighing approximately 2 kg were used. The rabbits were randomly divided into 4 groups. The bone segment was removed from the mid radial bone. In the first group (N = 5) in the gap segement of bovine rib cartilage was implanted. In the second group (N = 5) in the gap, segement of rabbit rib cartilage was implanted. In the third group (empty control group), (N = 5) the defect was left without implantation. Finally, the fourth group (autograft Group), (N = 5) the defect was filled with a same harvested bone. The skin and muscles were sutured routinely. X-rays were taken on 14th , 28th , 42nd and 56th postoperative days.

Result and conclusion

After 8 weeks bone samples were taken from healed area for for histopathological evaluation. The results of our study indicate allogenic and xenogenic cartilage acted almost like autograft groups and were better than empty group.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P067

Fabrication of porous and multifunctional composite materials for bone regeneration

*K. Besecke[1], N. Ehlert[1], P. Behrens[1], A. Hoffmann[2], L. Burmeister[2]
[1]Leibniz Universität Hannover, Institut für Anorganische Chemie , Hannover, Deutschland
[2]Medizinische Hochschule Hannover, Klinik für Orthopädie , Hannover, Deutschland

Currently, large bone voids over critical size are a clinical challenge.[1] The gold standard for the treatment are autografts, however, their use can lead to complications such as infections or donor-site morbidity thus synthetic alternatives are required.[2]

Our approach is to build up porous and multifunctional composite materials which should temporally replace the function of lost bone tissue and support the natural regeneration process. Bioactive glasses have emerged as promising candidates for the fabrication of porous bone substitute scaffolds. These materials are osteoconductive and possess an osteoinductive character.[3] The mechanical properties of this scaffolds can be enhanced with polymer coatings and the resulting composite materials should combine the qualities of the respective components.[4] For example, the biopolymer chitosan is suitable for stabilizing due to inherent biocompatibility and antibacterial properties.[5] Furthermore, the osteoinductivity of composites can be increased by the attachment of specific growth factors (Bone Morphogenetic Proteins, BMPs). Especially BMP-2 is the most studied representative of this group and is considered to be an effective growth factor that can induce bone regeneration.[6]

Macro- and nanoporous bioactive glass scaffolds were prepared via a (polyurethane) sponge templating technique using Pluronic®F127 as co-template to build up a nanoporous system. After calcination the template free scaffolds were coated with chitosan. Polymer contents were determined with thermogravimetry and C/S-analyses and the improved mechanical properties of composite scaffolds were analyzed by compressive strength measurements. BMP-2 was attached to the surface and the amounts were determined by an Enzyme Linked Immunosorbent Assay (ELISA).

The uncoated scaffolds had an interconnected pore network with macropores in a range of 100−500 µm, nanopores had a diameter of 6 nm. Chitosan coated scaffolds showed improved mechanical properties. Besides, the attachment of BMP-2 was successful.

References

[1] J.J. Li *et al*., *Journal of Materials Chemistry B*, **2014**, 2, 7272–7306.
[2] L. Polo-Corrales *et al*., *Journal of Nanoscience and Nanotechnology*, **2014**, 14, 15–56.
[3] M.M. Stevens, *materialstoday*, **2008**, 11, 18–25.
[4] J.R. Jones, *Acta Biomaterialia*, **2013**, 9, 4457–4486.
[5] M.S. Shoichet, *Macromolecules*, **2010**, 43, 581–591.
[6] T. Müller *et al*., *BIOspektrum*, **2007**, 3 236–240.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P068

Complementary reinforced magnesium phosphate cement

*T. Brückner[1], A. Stengele[1], J. Groll[1], U. Gbureck[1]
[1]Universitätsklinikum Würzburg, Abteilung für Funktionswerkstoffe der Medizin und der Zahnheilkunde , Würzburg, Deutschland

Introduction

Mineral bone cements such as calcium phosphate (CPC) or magnesium phosphate cements (MPC) suffer from brittle mechanical behaviour. Supplementing the cement with poly(acrylic acid) [PAA, $(C_3H_4O_2)_n$] is a common approach to circumvent ceramic brittleness in CPC research [2-4]. In the present study, we combined PAA with classic MPC formulations to pool a less brittle fracture behaviour with the biological advantages of MPC such as a higher biodegradation potential and the incorporation of osteogenic magnesium ions [1].

Methods

Farringtonite [$Mg_3(PO_4)_2$] was mixed with PAA (M_w=100,000 g/mol) solution, $(NH_4)_2PO_4$ solution or adequate mixtures of both at different powder-to-liquid-ratios (1.0-3.0 g/mL). The concentrations of the liquid phases varied between 0 to 35 % (PAA) and 0 to 3.5 mol/L [$(NH_4)_2PO_4$)], respectively. In dependency of the cement composition used, the behaviour while setting (pH, temperature), the workability (injectability), phase composition *via* X-ray diffractometry, surface morphology *via* scanning electron microscopy and wet strength under compression after 24 h at 37 °C were analysed.

Results

PAA alone was not suitable for the fabrication of mechanically stable cement matrices, as compressive strengths of 5 MPa were observed at most. Equally, low concentrations of $(NH_4)_2PO_4$ in the solution (≤1.75 mol/L) resulted in maximum 12 MPa. In contrast, using mixtures of both, a complementary effect from combined chelation and dissolution/precipitation reaction enabled a at least 3-fold reinforcement compared to the corresponding controls as well as a less brittle fracture behaviour. The cementitious reaction led to newberyite ($MgHPO_4 \cdot 3H_2O$) and struvite ($MgNH_4PO_4 \cdot 6H_2O$) as hydration products at which the amounts depended on the PAA and $(NH_4)_2PO_4$ amount in the liquid phase.

Conclusions

Indeed, we were not successful in enhancing the mechanical properties with PAA solely, but integration of standard reactants from MPC research resulted in workable cements with outstanding mechanical properties by synergistic reinforcement effects.

References

[1] N. Ostrowski, A. Roy, P. Kumta, *ACS Biomater. Sci. Eng.* 2016;2: 1067 - 1083.
[2] W. Chen, C. Ju, J. Wang, *Dent. Mater* 2008;24: 1616 - 1622.
[3] A. Majekodumni, S. Deb, *J. Mater. Sci.-Mater. Med.* 2007;18: 1883 -1888.
[4] A. Majekodumni, S. Deb, J. Nicholson, *J. Mater. Sci.-Mater. Med.* 2003;14: 747 - 752.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P069

Modified surfaces of advanced NiTi alloys in the in-vivo model of osteoblasts

*J. Sevcikova[1], A. Dlouhy[2], A. Weiser[2], P. Stepka[1], K. Vrchovecka[1], M. Pavkova Goldbergova[1]
[1]Masaryk University, , Brno, Tschechien
[2]Academy of Sciences of the Czech Republic, , Brno, Tschechien

The project focuses on materials issues associated with the in-vivo performance of advanced NiTi alloys with surface modifications (He, H, nanotubes) and the functional fatigue and the in-vivo environmental attack.

The biological stress and the cell response to the material (NiTi discs of 12-mm diameter and 1-mm thick) were evaluated in the cell culture media and on the human osteoblasts (HOb) in selected time periods (48 and 72 hours) under physiological and pathological conditions (osteoarthrotic HOb). For the measurement of Ni and Ti the ICP MS (inductively coupled plasma mass spectrometer), and PhenGreen dying method were used. Fluorescent microscopy and SEM was used to determine cell morphology and activity.

Regarding the material in the media solution, a relatively high concentration of Ni was observed after 24 hours with decrease in further periods, especially in He surface compared to H surface alloy. The adhesion, proliferation and viability were proved in all types of material (He, H, nanotubes). In the cell focused analysis, the analyses of metal in the cells revealed that Ni and Ti ions release is different with the lowest levels in He alloy. Different behaviour of osteoarthrotic osteoblasts compared to physiological HOb regarding the contact with the metal alloy was observed.

In conclusion, the ion release, which could be related to passivation, damage and corrosion of the material, is affects the cell behaviour, not only in the proliferation but also in the energetic activity of the cells.

The study was supported by research project GA15-16336S of the Grant agency of the Czech Republic.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P070

Detection of Strontium release from two new composite bone graft substitute materials with differing amounts of strontium

*N. Döhner[1], C. Kern[2], S. Ray[3], U. Thormann[3], V. Alt[3], B. Kruppke[1], T. Hanke[1], M. Rohnke[2], J. Janek[2], L. Franz-Forsthoffer[3]
[1]Technische Universität Dresden, Max Bergmann Center of biomaterials Dresden , Dresden, Deutschland
[2]Justus-Liebig-University of Gießen, Institute for Physical Chemistry , Gießen, Deutschland
[3]Justus-Liebig-University of Gießen, Laboratory of Experimental Trauma Surgery , Gießen, Deutschland

Osteoporosis is a severe bone disease leading to loss of bone mass and thereby decreased load bearing capabilities. The need for novel bone graft substitutes for locally delivering a better and faster healing to these pre-damaged regions is of high importance, because osteosynthesis often fails to stabilize this special type of fractures.

In this study two new bone graft substitute materials are tested, PPGC+S III 5:5 (5:5) and PPGC+S III 3:7 (3:7), respectively. The materials were phosphate **p**restructured gelatin mineralized with **c**alcium and **s**trontium, which is supplemented by the Ca/Sr ratio (5:5 and 3:7, respectively). Both biomaterials contain strontium, which enhances bone formation on one hand and decreases bone resorption on other. *In vitro* tests conducted demonstrate the composite ability to stimulate osteoblast proliferation. In addition, the materials show good porosity, osteo-conduction and biocompatibility.

24 female Sprague-Dawley rats were randomized into 3 groups (n=10 for empty control, n=6 for (5:5), n=8 for (3:7)). A combinatorial approach of multi-deficiency diet for 3 months after bilateral ovariectomy was used for induction of osteoporosis. The left femur underwent a 4mm wedge-shaped metaphyseal osteotomy that was internally fixed with a T-shaped plate. The defect was then either filled with PPGGC+S III 5:5 or 3:7 and internally stabilized with a T shaped mini-plate. After 6 weeks femora were harvested followed by Micro-CT, histological, histomorphometrical and enzymehistochistochemical (ALP; TRAP) analysis.

Time of flight secondary ion mass spectrometry technology was used to assess the distribution of released strontium ions and calcium appearance of newly formed bone. Histomorphometric analysis showed a statistically significant increase in the bone formation and osteoid formation in PPGC+S III 3:7 ($p<0.01$). This was further validated by Micro Ct data. In addition an increased ALP activity was also seen in 3:7. Although material degradation was seen in both groups interestingly, increased TRAP activity was also seen in case of 3:7. TOF-SIMs analysis showed a higher release of Sr from PPGC+S III 3:7 into the interface region and related to a higher calcium content in this area compared to PPGC+S III 5:5.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P071

Development of composed material based on zirconium oxide

*I. Lytkin[1], A. Buyakov[1], I. Kurzina[1]
[1]National Reseach Tomsk State University, , Tomsk, Russische Föderation

A number of composite materials for the restoration of bone tissue based on zirconium oxide and lactide are obtained. The method of XRD evaluates the composition of materials at different stages of production [1-2]. It is established that during the sintering of zirconium oxide a baddeleyite phase is formed, which remains unchanged throughout the entire cycle [3]. The quantitative estimation of micro- and macroporosity of materials is given. A simplified step-by-step production scheme is shown in Fig. 1.

Fig. 1 - Scheme for producing a composite material

Photomicrographs of RSMA Fig. 2a show the distribution of the phases of zirconium, magnesium and oxygen over the surface. To the left, the original zirconium ceramics immediately after sintering, on the right after the composite, after 24 hours of impregnation with polylactide. Scanning electron microscopy was done to determine the distribution of pores on the ceramic surface, for a more detailed understanding of the structure of the composite material. Fig. 2b presents microphotographs of the surface of zirconium ceramics without a pore formation (right) and with a pore formation agent (on the left), on which a uniform distribution of pores is observed over the entire surface of the material.

Fig. 2 a – The distribution of particles on the X-ray spectrograph (1 - initial zirconium ceramics, 2 - Composite material 24 hours after impregnation)

b - Microphotographs of the surface of zirconium ceramics (1 - without pore formation agent, 2 - with pore formation agent)

Materials based on zirconium oxide and polylactide are obtained. The presence of the ZrO2 phase by X-ray phase analysis was confirmed. The role of the porosity and duration of interaction with the polymer solution on the amount of polymer material adsorbed in the porous space of the ceramic material is established. As the contact time with the polymer solution increases, the volume porosity of the materials decreases proportionally. For materials without blowing agent by 50% and for materials with pore-forming agent by 30%. The maximum amount of the introduced polylactide is observed on samples without a pore former.

[1] V. Sollazzo, F. Pezzetti, A. Scarano, A. Piattelli, C.A. Bignozzi, L. Massari, G. Brunelli, dental materials 24, 357–361 (2008).
[2] S. Saravanan, R.S. Leena, N. Selvamurugan, International Journal of Biological Macromolecules, 123-126 (2016)
[3] R. Tejero, E. Anitua, G. Orive, Progress in Polymer Science 1 - 144 (2014).

Figure 1

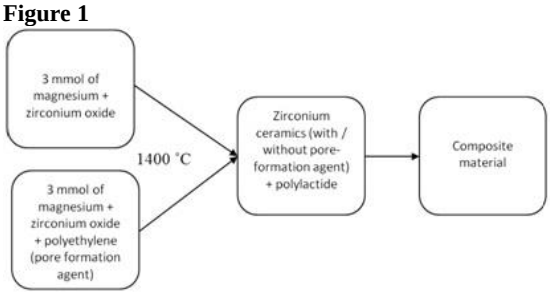
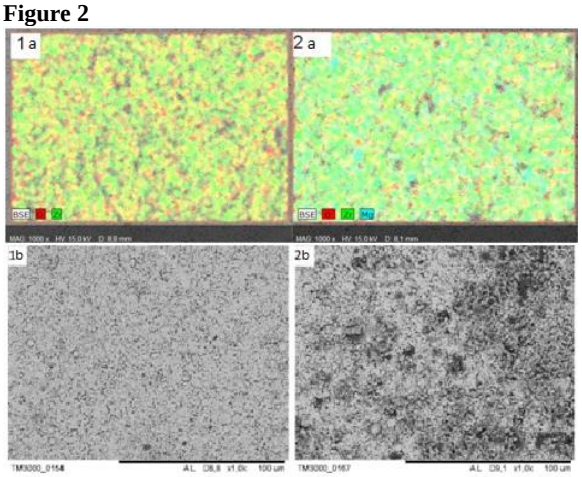


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P072

3D Powder Printing of Strontium-Magnesium Phosphate bone implants and the impact of the chemical composition on bone cells

S. Meininger[1], K. Spatz[1], E. März[1], A. Ewald[1], J. Groll[1], *E. Vorndran[1], U. Gbureck[1]
[1]University Clinic Würzburg, Department for Functional Materials in Medicine and Dentistry , Würzburg, Deutschland

Introduction

3D powder printing of biocompatible materials is an excellent method for the fabrication of individual bone grafts with complex geometry. The use of biodegradable materials with a similar chemical composition to bone and a porous structure provides optimal conditions for bone ingrowth resulting in strong implant integration. The suitability of additive manufactured calcium phosphate cements (CaP) as individual bone implants was already proven [1]. A promising alternative is given by magnesium phosphate cement (MgP), due to its biocompatibility and faster biodegradation [2]. In this study MgP was modified with strontium (Sr), in order to increase radiopacity of the implant and reduce osteoclast proliferation.

Methods

Cements of $Mg_{3-x}Sr_x(PO_4)_2$ (x = 0-1) was powder printed (Z510, Z-Corp) with a 1M $NH_4H_2PO_4$ solution. The radiopacity, compressive strength, chemical composition (XRD, EDX), porosity, microstructure (SEM), and Sr release (ICP-MS), were analysed. The osteogenetic potential of Sr-modified MgPCs was evaluated in cell culture with MG63 cells or RAW264.7 cells.

Results

The solidification of the cements was based on the conversion of $Mg_3(PO_4)_2$ to struvite ($MgNH_4PO_4 \cdot 6H_2O$); while the Sr-containing phases was unreactive and existed as intermediate crystals within the struvite matrix. The specimen showed a high porosity of (21±1) %, which increased by the addition of Sr (29±1) %. This results in a decrease of compressive strength with rising Sr content from 20 MPa to 12 MPa. Compared to unmodified MgP the radiopacity was enhanced 4 times by the addition of Sr. Sr-release was independent to Sr content (25–40 µM/d) and effected the osteogenic potential of MgP (Fig.2). Particularly, it resulted in an increased osteoblast activity and showed similar cell numbers compared with osteoblasts grown on CaP. The cell number and activity of osteoclasts were significantly reduced by the addition of Sr.

Conclusion

Highly accurate MgP structures were printable (Fig.1). The *in vitro* cell compatibility could be enhanced by Sr-modification enhanced, which promises a high osteogenetic potential *in vivo*.

Fig. 1: 3D printed MgPs.
Fig.2: A) osteoblast activity, B) osteoclast cell number.

References

[1] Klammert U. et al., J Cranio Maxill Surg 2010; 38: 565
[2] Kanter B. et al., Acta Biomaterialia 2014; 10: 3279

Figure 1

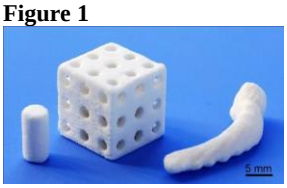
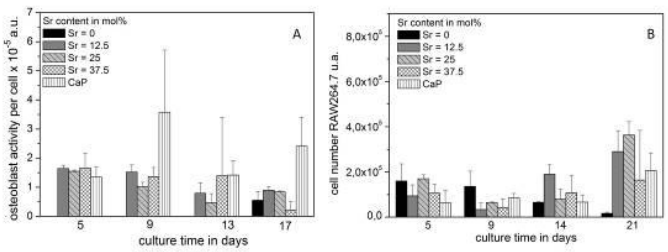


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P073

Biocompatibility test of the TiNb adhesion interlayer for bioactive coatings

*M. Parizek[1], Z. Tolde[2], V. Stary[2], L. Cvrcek[2], M. Vandrovcova[1], J. Remsa[3], S. Danis[4], J. Krcil[2], L. Bacakova[1], P. Spatenka[2]
[1]Institute of Physiology, Dept. of Biomaterials and Tissue Engineering , Prague, Tschechien
[2]CTU in Prague, Dept. of Materials Engineering, Faculty of Mechanical Engineering , Prague, Tschechien
[3]CTU in Prague, Faculty of Biomedical Engineering , Kladno, Tschechien
[4]Charles Univ. in Prague, Dept. of Cond. Matter Physics, Fac. of Math. and Phys. , Prague, Tschechien

The aim of our present work was to compare b-TiNb alloy films prepared on various bulk substrates for use in bioapplications. TiNb alloy in the form of a coating is targeted for use as barier and adhesion interlayer, especially as a substrate for an electrically active BaTiO3 layer, so the human cell growth and the toxicity of this material system must be tested.

Four types of materials with identically prepared surfaces were used for sputtering of the TiNb layer: Ground and polished coupons of β-Ti39Nb, cpTi ISO 5832-2 (cpTi grade2), Ti alloy Ti6Al4V ISO 5832-3 and austenitic stainless steel AISI 316L ISO 5832-1 were used as substrates. In what follows, the substrates are usually denoted as TiNb, Ti, TiAlV and Fe (steel), respectively.

The TiNb layers were prepared by magnetron sputtering in a Flexicoat 850 (Hauzer, Netherland) coating machine on prepared substrates. The thickness of the TiNb layer was measured with a Calotest (CSM, Switzerland), and was found to be 2.4+/-0.1 µm.

To test the biological performance of used materials we prepared the BaTiO3 layers on Ti and TiNb substrates. The layers were prepared by the pulsed laser deposition method (PLD) .

Ti and TiNb covered with BaTiO3 samples were sterilized with 70% ethanol, inserted into 24-well polystyrene cell culture plates (PS; TPP, internal well diameter 15.6 mm) and were seeded with human osteoblast-like Saos-2 cells (ECCC, Salisbury, UK). The cells were cultured for 1 and 3 days (to evaluate the number of cells and viability) or for 7 days (to evaluate the number of cells) at 37 °C in a humidified air atmosphere containing 5% CO2.

The test of cell growth confirmed that the BaTiO3 layers were non-toxic and suitable for cell growth. The Saos-2 cells grown on all BaTiO3 surfaces were highly viable (97-99%). The cell density on BaTiO3 films on the TiNb substrate was higher even after one day of cultivation then on Ti substrate.The positive influence of Nb can be connected with a chance of some cells to find surface uncovered by BaTiO3 film or in existence of some mechanism of transport of Nb ions through this film.

Moreover, the TiNb can be a promising substrate for active layers such as BaTiO3 with very appropriate characteristics for medical applications. It was found the combinations both Ti/BaTiO3 and TiNb/BaTiO3 are biocompatible and non-toxic.

*Supported by the Grant Agency of the Czech Republic (grant No. **15-01558S**).*

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P074

Composite materials based on calcium phosphates and poly(vinyl alcohol) for bone reconstruction

*P. Kalachikova[1], D. Lytkina[1], I. Kurzina[1]
[1]National Reseach Tomsk State University, , Tomsk, Russische Föderation

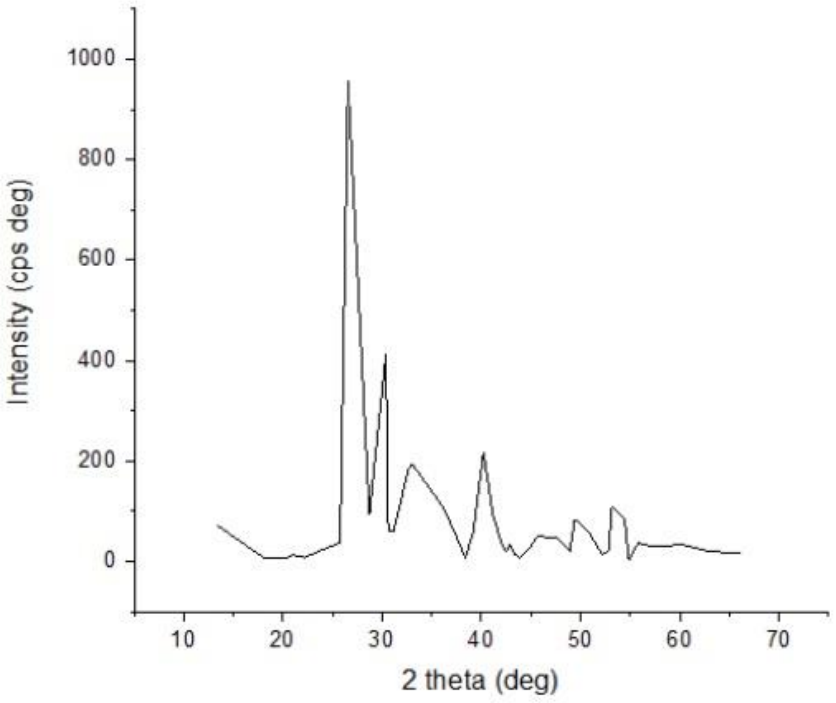
Bone is known as a natural-organic composite material of complex structure. Basically, it consists of nano-scale inorganic crystals that filter collagen fibrils and water. Synthesized hydroxyapatite (HA) is of scientific interest due to biocompability and osteoconductive properties. Besides, being non-toxic and biodegradable, hydroxyapatite is suitable for biomedical materials. In recent years, particular attention has been paid to the preparation of HA bioceramics with porous morphology. Porous HA exhibits strong bonding to the bone; the pores provide mechanical interlock leading to material firm fixation. Another polymer with potential biomedical application is poly(vinyl alcohol) (PVA). PVA is a polar hydrophilic biocompatible polymer possessing excellent physical and mechanical properties of absorption and exudation of body fluid; which has led to its applications as a biomaterial.

The primary target of the current paper is to describe synthesis of poly(vinyl alcohol) – hydroxyapatite hydrogels and composites with different HA and PVA ratios, as well as the characteristics of obtained compounds. HA – PVA composite materials were obtained in forms of both hydrogel and powders with different ratio of components by means of adding hydroxyapatite powder to PVA water solution. Another synthetic route was carried out by mixing reagents for hydroxyapatite with PVA water solution on continuous heating and stirring throughout eight-hour period. In both methods suspensions were washed with distilled water, dried, and divided in halves – whereas one half was calcined with subsequent white powders obtainment, and another half was being processed through several freezing-thawing cycles with subsequent hydrogels obtainment.

XRD results have indicated that a variety of calcium phosphates – such as monetite ($Ca(HPO_4)$) and brushite ($Ca(HPO_4)\cdot 2H_2O$) – was formed in the samples prepared by mixing reagents for hydroxyapatite and PVA, and hydroxyapatite phase was not indicated in neither of the samples. Still, the samples appeared to have well-crystallized structure. Composites obtained by mixing HA with PVA are more amorphous, yet, they do contain hydroxyapatite crystalline phase.

Fig. 1. XRD-graph of Monentite – PVA composite

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P075

Strontium functionalization of Ti-40Nb implants by reactive sputtering

M. Göttlicher[1], *M. Rohnke[1], Y. Moryson[1], M. Schumacher[2], J. Thomas[3], J. Sann[1], T. Gemming[3], R. Schmidt[3], S. Pilz[3], A. Gebert[3], A. Lode[2], J. Janek[1]
[1]Justus-Liebig-Universität Gießen, Physikalische Chemie , Gießen, Deutschland
[2]Technische Universität Dresden, Centre for Translational Bone, Joint and Soft Tissue ResearchCentre for Translational Bone, Joint and Soft Tissue Research , Dresden, Deutschland
[3]IFW Dresden, Institute for complex materials , Dresden, Deutschland

Implant surface modification and functionalization is a modern approach to enhance the osseointegration of bone implants, especially in the presence of systemic bone diseases like osteoporosis. Here we developed a Sr containing coating for Ti-40Nb alloys. Sr^{2+} is a therapeutic agent, which stimulates bone growth. Instead of an oral administration, a controlled and localized strontium release is achieved with the aim to minimize side effects and enhance osseointegration. Titanium-niobium was designed as a special implant alloy with advanced mechanical properties in comparison to conventional titanium implants.

The strontium containing thin films were prepared by reactive sputtering in a 13.56 MHz cpacitively coupled radio frequency plasma reactor by the use of a strontium chloride target. The structure and composition of the sputtered films were investigated by scanning electron microscopy (SEM), time of flight secondary ion mass spectrometry (ToF-SIMS), high resolution transmission electron microscopy (HR-TEM), X-ray photoelectron spectroscopy (XPS) and energy dispersive X-ray spectroscopy (EDX). The bioactivity of the surface was determined by cell cultivation of human osteoblasts and subsequent analysis of cell morphology, viability, proliferation and differentiation. The strontium release was also controlled in TBS via inductively coupled plasma mass spectrometry (ICP-MS).

Dense coatings with a thickness of more than 100 nm could be produced. The coatings consist of a mixed metal oxide halogenide as shown by XPS and EDX. HR-TEM measurements of the cross section indicate an incoherent but well-structured interface between the coating and titanium-niobium substrate. Cell studies show a significant improved cell activation and cultivation in comparison to uncoated implants. The released amount of strontium is close to the therapeutic dose.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P076

In vitro degradation of novel medical polyurethanes. morphology and mechanical characterization

*K. Athanasopulu[1], *L. Kutuzova[1,2], M. Schneider[2], S. Sun[2], R. Kemkemer[1,3], G. Lorenz[1]
[1]Hochschule Reutlingen, Angewandte Chemie , Reutlingen, Deutschland
[2]Reutlingen Research Institute, Polymere , Reutlingen, Deutschland
[3]Max-Planck-Institute for Intelligent Systems, , Stuttgart, Deutschland

The initial estimate of bioperformance of the polymer-based formulations in medical devices can be realized by rapid and inexpensive in vitro investigations to simulate in vivo physiological conditions. Biostability of chondral and osteochondral implants made of polymers is strongly influenced by mechanical stress and reactive oxidative intermediates in physiologic environment of the joints. On long time scales oxidative degradation and environmental stress cracking leads to device failure and reduces life time of implants [1,2].Thermoplastic polycarbonateurethane (TPCU) elastomer with E-Modulus lower than 15 MPa seems to be an interesting material calls for chondral and osteochondral implants. Elastomeric materials mimic the mechanical response of the natural meniscus without damaging the interconnected articular cartilage during compressive load. Furthermore, these materials exhibit better oxidative resistance compared to polyether urethane as well as an excellent wear resistance if a soft segment includes chemical bounded silicone lubricant [3]. Composition variations in segmented TPCUs strongly affect degree of segment ordering as well as degree of phase separation and can lead to significant variations in degradation kinetics. For predicting TPCU biostability in synovial fluids the accelerated metal ion-catalyzed method has been recommended to simulate in vivo degradation in unloaded regions with acceleration ratio of 1:15 [3]. The aim of this study was to screen the oxidative resistance of a series of novel as well as commercially available biomedical TPCUs to predict their aging in vivo. The adequate biostability of the materials based on TPCU-PDMS-blockcopolymers was achieved varying the structure parameters. All TPCU-materials have been developed to ensure application relevant mechanical properties and to have a favorable bio performance under continuous physiological loadings. Degradation profiles have been monitored using FTIR to verify changes in molecular structure, SEM to observe surface changes, DSC and DMA to monitor changes in thermal and mechanical properties and GPC to detect changes in MW distribution (Fig.1-2). New formulations of soft TPCUs exhibit the valid bio-performance after 5 months using an accelerated testing method.

References

[1] Christenson, E.M. *at al., BiomedMaterResA* 70:245-255, 2004
[2] Zhao, Q *et.al., J.BiomedMaterRes* 29:467–475, 1995
[3] Dempsey, D.K. *et.al., J.BiomedMaterRes* PartA 102A:3649-3665,2014

Figure 1

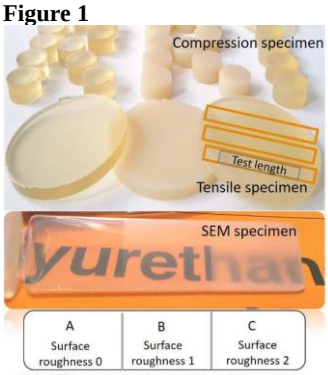


Fig 1. TPCU- specimens to control the surface and bulk properties in storage experiments

Figure 2

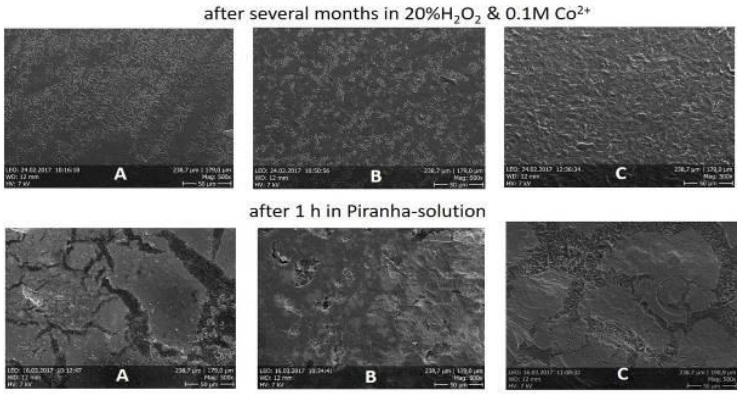


Fig 2. Storage experiments with oxidizers. Effect of the surface roughness

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P077

Stable immobilization of proteins and peptides on bioinert high-performance ceramics for the promotion of cell adhesion, migration, and differentiation

*F. Böke[1], N. Labude[2], I. Lauria[1], S. Neuss[2,3], H. Fischer[1]
[1]RWTH Aachen University, Dental Materials and Biomaterials Research , Aachen, Deutschland
[2]RWTH Aachen University, Institute of Pathology , Aachen, Deutschland
[3]RWTH Aachen University, Helmholtz Institute for Biomedical Engineering - Biointerface Group , Aachen, Deutschland

High-performance oxide ceramics have successfully been used as medical implants for decades due to their high strength, high wear resistance and cytocompatibility. However, these ceramics are not suitable for direct bone contact due to an insufficient osseointegration. To enable a broader use in clinical practice, their bioinert surfaces need to be modified to promote cell recruitment and subsequent tissue integration. Therefore, we have developed a novel strategy to efficiently bioactivate oxide high-performance ceramic surfaces through tailored, stable silicate/silane coatings for peptide and protein coupling.

High-performance oxide ceramics were coated with a 100 nm thick SiOx-layer via physical vapor deposition. Afterwards, a silane monolayer was applied to present either –NH2, -SH, or a mixture of both groups at the interface. The application process was verified via XPS, FTIR-ATR, EDX, and AFM. After silanisation, the –NH2 and –SH group density was calculated at > 90 % on the mono-functionalized and > 80% on bi-functionalized surfaces. AFM and XPS analyses confirmed a silane monolayer of approximately 0.7 nm thickness.

Coupling efficiency of different peptides and proteins was proven by various methods: for RGD-peptides by I125-radioactive labeling, for HGF and BMP-2 by AuNP-labeling. To analyze the retained function of RGD after coupling, we developed a custom centrifugation-assay with murine fibroblasts (L929) and human mesenchymal stem cells (MSC). Potential loss-of-function of the HGF after coupling was examined by MSC and HuH7 Boyden chamber assays. Osteogenic induction potential of BMP-2 functionalized and unmodified ceramics was evaluated by RealTime-PCR.

I125-labeling shows significant amounts of RGD when crosslinked on the surface in comparison to unspecific binding. A significant higher adhesion of L929 and MSC on RGD-loaded ceramics compared to RAD-loaded substrates was determined via centrifugation-assay. AuNP-labeling confirmed the accessibility of BMP-2 and HGF after crosslinking. Migration studies of HGF after coupling showed significant higher amounts of cells migrating towards the modified surfaces. RT-PCR showed significant higher osteogenic differentiation of MSC on BMP-2 coupled surfaces.

In conclusion, individual chemical functionalization enables directed immobilization of cell adhesion, migration and differentiation promoting peptides and proteins to significantly enhance osseointegration on high-performance oxide ceramics.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P078

Analysis of the spinning process and batch-to-batch consistency of aligned PCL/Collagen nanofibers for skeletal muscle tissue

*D. Dippold[1], D. W. Schubert[1], J. P. Beier[2], A. R. Boccaccini[3]
[1]Friedrich-Alexander Universität Erlangen Nürnberg, Lehrstuhl für Polymerwerkstoffe , Erlangen, Deutschland
[2]Uniklinik Erlangen, Plastische- und Handchirurgie , Erlangen, Deutschland
[3]Friedrich-Alexander Universität Erlangen Nürnberg, Lehrstuhl für Biomaterialien , Erlangen, Deutschland

The application of tissue engineered constructs is a promising approach in regenerative therapies for damaged skeletal muscle tissue. It aims to deliver healthy and functional cells on a mechanically supporting matrix to the damaged area of the tissue. Moreover, aligned and patterned topographies can guide cell alignment and support the formation of aligned myotubes. Therefore, the development of highly aligned scaffolds based on polycaprolactone (PCL) has been studied extensively [1]. In this study, aligned polycaprolactone/collagen (PCL/Col) biocomposite nanofibers were fabricated via electrospinning using environmentally benign acetic acid as solvent. Acetic acid was used only once with the PCL/Col material combination, resulting in very inhomogeneous fibers and bead formation [2]. Therefore the solubility and spinnability of different PCL/Col ratios and different collagen batches were investigated. The most suitable solutions were spun at various spinning parameters and collected with different collector setups in order to determine ideal processing conditions for the fabrication of highly aligned nanofibers. Light microscopy and SEM were used to investigate the fiber morphology, yield and alignment. The results demonstrate that ultrasonic treatment helps to fully dissolve PCL and that it is possible to fabricate homogenous PCL/Col fibers. Nanoscale fiber diameters can be achieved, depending on the process parameters. Additionally, various parameter fields were investigated in order to determine the most stable and suitable parameters for nanofiber fabrication [3]. In the final step, multiple collagen batches very analyzed according to their viscosity and spinnability in order to investigate the challenging, intrinsic batch-to-batch inconsistency of natural polymers.

References

[1] Doergens, A.; Materials Letters, 2015, 140, 99.
[2] Yogeshwar, C.; Journal of Applied Polymer Science, 2012, 125, 3221.
[3] Dippold, D.; Materials Science and Engineering C, 2017, 72.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P079

Dispersing Heterotopic Ossification with Hexametaphosphate

*T. Robinson[1,2], N. Eisenstein[1,2], S. C. Cox[1], S. Stapley[2], L. M. Grover[1]
[1]University of Birmingham, School of Chemical Engineering , Birmingham, Vereinigtes Königreich
[2]Royal Centre for Defence Medicine, , Birmingham, Vereinigtes Königreich

Introduction

Heterotopic ossification (HO) is the pathological formation of ectopic bone, and has become an epidemic in the military following blast trauma. HO can cause chronic pain and skin ulceration, and creates difficulties with prosthetic limb fitting. Current prophylaxes are suboptimal and are associated with unacceptable side effects. Once formed the only treatment for HO is excision surgery.

Objectives

The aim of this work is to develop a minimally invasive treatment and prophylactic for HO, utilising hexametaphosphate (HMP) as the active therapeutic. HMP is a potent calcium chelator, and has been shown to prevent calcium phosphate crystallisation *in vitro*, and demineralise bone *ex vivo*. The objectives of this study are to load HMP into a suitable vehicle for local drug delivery, such as an injectable or small implant, and test the material and HMP release properties *in vitro*, and the safety and efficacy *in vivo*.

Materials and Methods

HMP was incorporated, in solution concentrations of 0 – 1 M, into hydrogel polymer systems, including non-cross-linked sodium alginate, between 1 – 10 w/v%. The material properties of these systems were analysed by rheology and injectability testing. The HMP elution from these systems was tested with a release assay.

HO was induced in the lower legs of rodents by performing Achilles tenotomy surgery. This model was used to test the efficacy of HMP formulations to prevent HO, as well as examining any deleterious effects. The formed bone volume was measured using micro computed tomography.

Results

HMP may be loaded into viscous alginate solutions. Rotational rheology shows an increase in viscosity with HMP concentration up to 0.2 M, and a decrease in viscosity above this concentration, consistent with observed precipitate formation. This suggests cross-linking between HMP and the polymer, likely via sodium ions present. Solutions are shear thinning, which is advantageous for injectable systems.

A decrease in formed HO volume is seen in animals which received HMP, compared to those that received blank controls. No deleterious side effects of using HMP *in vivo* have been detected thus far.

Conclusion

HMP is encouraging for use as a therapeutic to treat HO, however its activity must be localised to the site of incidence. Sodium alginate solution is a promising carrier, though further characterisation is of this system is required for optimisation. The animal model has shown HMP to be both safe and efficacious.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P080

Biostable polyurethane materials for chondral implants: in-situ monitoring of urethane polymerization

*L. Kutuzova[1,2], O. Rain[2], R. Koslik[2], G. Lorenz[1], A. Kandelbauer[1]
[1]Reutlingen University, School of Applied Chemistry , Reutlingen, Deutschland
[2]Reutlingen Research Institute, School of Applied Chemistry , Reutlingen, Deutschland

Thermoplastic polycarbonate-urethane (TPCU) elastomers are currently attracting great interest as a promising basic matrix for fabricating the next generation of biostable, implantable biomedical devices [1]. Segmented polyurethane block co-polymer chains consist of alternate soft and hard segments, each of which contains up to 5 repeating units in case of the hard segments and up to 20 repeating units in case of the soft segments. Variations in block lengths, polarity and connecting (coupling) order of the hard and soft segments in a polymer chain lead to different chemical compatibility between the structural elements and introduce wide changes in block-co-polymer morphology, thermo-mechanical behavior as well as hydrolytic stability and biocompatibility of the potential medical materials [2]. Hence, it is extremely important to control co-polymer formation and segmental constitution during synthesis. Undefined variations in the synthesis protocol of high molecular TPCU block-copolymers will strongly affect the molecular structure of the polymer chains as well as their molecular weight distribution in step-growth polyaddition of diols and diisocyanates. The goal of this project was to develop catalyst-free, multi step synthesis procedures for optimizing the large scale production of medical-grade TPCU elastomers with controlled molecular structure and physical-mechanical properties.

A series of the biomedical TPCUs with different percentages of hard segment and a silicone component in the soft segment were synthesized (Fig.1). The kinetic profiles of the urethane formation in polycarbonate-silicone-urethane-urea-block copolymer systems were monitored by in line FTIR ATR spectroscopic (React IR) and real-time calorimetric (RC1) methods (Fig.2). Moreover, the low molecular weight fractions were characterized off-line via NMR spectroscopic, rheological and thermomechanical methods. Mechanic response of the elastomer end-products to cyclic compression and tensile loading were systematically studied to be used to control the reaction reproducibility.

Acknowledgement: Financial support of this project by BMBF under grant number 01EC1406C (TOKMIS) as well as by GEPRIS.DFG project 253160297 are gratefully acknowledged.

References

[1] *Comprehensive Biomaterials.* P.Ducheyne et al, Elsevier Sci, 2011, 3672 p
[2] *Polyurethane Elastomers. From Morphology to Mechanical Aspects.* C.Prisacariu, Springer, 2011, 255 p

Figure 1

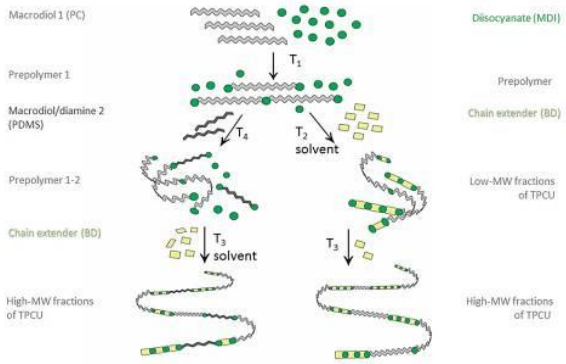


Figure 1. Designing *multistep* one-pot TPCU-polymerization

Figure 2

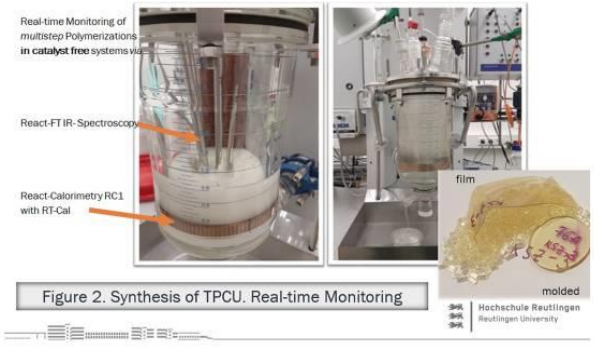


Figure 2. Synthesis of TPCU. Real-time Monitoring

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P081

Patterns from biocompatible polymer and native protein for organizing cells in tissue like fashion

*G. Pilarczyk[1], M. Gunkel[2], L. Keiber[1], H. Erfle[2], M. Hausmann[1]
[1]Kirchhoff Institut für Physik, F18 , Heidelberg, Deutschland
[2]Bioquant, , Heidelberg, Deutschland

The local environment of an individual cell inside an organized multicellular ensemble like a tissue patch or an organ is of paramount importance for cell integrity, development, activity and turnover. By interaction on basis of extracellular matrix (ECM) composition changes cells communicate with each other, thus making the local cell exterior an important component of organizing during tissue generation. Besides the biochemical function of ECM for synchronizing cell ensemble activities, mechanical and geometrical local cues imprint cell organizing features to generate tissue. We recapitulate such ECM properties to deposit geometrical and biochemical cues for cell organization during artificial tissue construction.

By a combined approach consisting of cell adhesion data analysis and surface patterning techniques a cell compatible substrate is decorated with geometrical pattern motives which organize large numbers of individual cells in a tissue imitating fashion. This way myoblasts of the cell line C2C12 are orientated line wise under conditions favoring cell division and proliferation. The resulting organized monolayer is stimulated to differentiate into muscle fibers while the underlying surface geometry pattern organizes the developing fibers as in the native muscle. Such surface patterns are done in a set of materials including siloxane (PDMS) but also poly-lactid acid (PLA).

The above described process exclusively uses possibilities given by pattern geometry. To further access the possibilities of ECM chemical cell stimulation parameters, a method for setup surface patterns in native protein is developed and applied to C2C12 cells. For this purpose the modification of serine side chain moieties by maleinimide coupling chemistry introduces soft UV-light dependent cross coupling acrylic groups into an ECM protein as collagen in the present case. The resulting modified protein is used to generate surface patterns from cell instructive protein material. The excellent bio-compatibility makes it possible to remove the modified protein by cellular phagocytosis during the cell organization procedure. As a result the geometrically organized multicellular ensemble replaces the primary artificial ECM by a secondary one of cellular origin.

Using differentiating stem cells may lead to artificial tissues with both a high definition of local differentiation and tissue like organization.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P083

An *in vitro* model of mature bone for pathological investigations

*A. Iordachescu[1,2], H. Amin[3], S. Rankin[3], R. Williams[1], C. Yapp[2,4], A. Bannerman[1], A. Pacureanu[5], O. Addison[6], P. Hulley[2], L. M. Grover[1]
[1]University of Birmingham, Chemical Engineering , Birmingham, Vereinigtes Königreich
[2]University of Oxford, Botnar Research Centre , Oxford, Vereinigtes Königreich
[3]Imperial College London, Inflammation, Development and Repair, National Heart & Lung Institute, Faculty of Medicine , London, Vereinigtes Königreich
[4]Harvard, Medical School , Boston, Vereinigte Staaten
[5]European Synchotron Radiation Facility, Beamline Groups Unit , Grenoble, Frankreich
[6]University of Birmingham, School of Dentistry , Birmingham, Vereinigtes Königreich

Bone is a tissue that continuously adapts to changes in mechanical load. This process can also result in maladaptive ectopic bone in response to injury and extreme mechanical insult, in a group of conditions known as heterotopic ossification.

Despite recent advancements, pathological differences at the molecular and structural levels are poorly understood. A number of *in vivo* models exist but can often be unreliable or too complex to allow isolation of factors which may stimulate progression of ossification.

We have developed a biologically-dynamic model of mature bone formation using a fibrin gel cast between two calcium phosphate ceramic anchors. These bioinspired early wound analogues are seeded with primary femoral periosteal cells - key players in a range of pathologies- and develop longitudinally over significant culture periods, allowing to study the temporal evolution of bone mineral and microstructure in excess of a year. We demonstrate the production of mature, bone-like morphology and chemistry, with differentiation of mature, mineral phase osteocytes.

Raman spectroscopy and XRD revealed that the mineral was bone hydroxyapatite and associated with collagen. Second harmonic imaging demonstrated that collagen was organized similarly to mature mouse femora. The initial stem cell population differentiated to the terminal osteocytic phase, was integrated into longitudinal networks similar to the canaliculi in mature bone (as demonstrated with nanoCT) and remained viable over the full year of culture. Using this model, we also demonstrated that pharmacological compounds can prevent the progress of ossification, displaying promise for applications in drug screening. We have also shown that it is possible to initiate angiogenesis in these constructs, with endothelial tubes also aligned with the mechanical axis, a step closer to mimicking *in vivo* bone.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P084

Multiscale hydrogel based structuring techniques for biomimetic hard tissue constructs.

D. Longhorn[1], T. Nedal[2], A. Molska[2], A. Håti[3], S. Bjørnøy[2], P. Sikorski[2], *D. Bassett[1,2]
[1]University of Birmingham, Chemical Engineering , Birmingham, Vereinigtes Königreich
[2]Norwegian University of Science and Technology (NTNU), Department of Physics , Trondheim, Norwegen
[3]École polytechnique fédérale de Lausanne (EPFL), Department of Materials , Lausanne, Schweiz

Cell based tissue engineering requires scaffolds that are both biofunctional i.e. provide the necessary environment for cell survival and maturation, and possess suitable mechanical and physicochemical properties for their application. In addition to tissue engineering applications, synthetic tissue-like constructs can be used as tools for 3D cell biology studies and for the purposes of materials and drug testing. If a suitable synthetic environment that closely mimics human tissue physiology can be created, then costly and ethically questionable *in vivo* animal studies or poorly relevant 2D cell culture tests could largely be avoided for the purposes of biomedical testing. Clearly such a tool would certainly accelerate progress in the field of regenerative medicine.

We have been working towards this goal in the context of bone tissue engineering by using a combination of soft materials structuring techniques, primarily using alginate, across a variety of length scales. Alginate hydrogels have a long and successful application in pharmacy, medicine and biomedical sciences and are routinely used to encapsulate various types of cells to provide a synthetic extracellular matrix (ECM), provide immunoprotection, or transport cells to the point of injury. However, significant material challenges arise from this approach, which are primarily derived from limitations in current physical structuring techniques and limited interaction with the cell membrane for unmodified alginates. These limitations present particular problems for certain biofabrication approaches and applying adherent cells respectively. This presentation will outline our developments in three key areas 1) Control of hydrogel gelation kinetics, 2) Patterning of cell architectures and 3) Synthesis and characterisation of mineralised architectures. Our focus is to create structures which can easily be interrogated *in situ* and some of our approaches to do this using a range of optical techniques will be discussed in particular.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P085

Synthesis and characterisation of Ga-doped ordered mesoporous bioactive glasses for biomedical applications

*F. E. Ciraldo[1], W. H. Goldmann[2], *A. R. Boccaccini[1]
[1]FAU Erlangen-Nürnberg, Lehrstuhl Biomaterialien , Erlangen, Deutschland
[2]FAU Erlangen-Nürnberg, Center for Medical Physics and Technology , Erlangen, Deutschland

Introduction

Since their discovery in the late 1960s, bioactive glasses (BGs) have been widely used for biomedical applications thanks to their ability to bond with hard and soft tissue once in contact with biological fluids. The development, in 2004, of a new family of glasses known as mesoporous bioactive glasses (MBGs) opened the possibility to overcome the limits of the traditional glasses produced by melt-quenching and sol-gel techniques. MBGs are characterised by high specific surface area, tunable mesoporosity with nanopores of 5 to 20 nm and by their fast biomineralization. These unique features make MBGs optimal candidates for the controlled delivery of drugs, biomolecules or therapeutic metallic ions. In this work, two compositions of Ga-doped MBGs with antibacterial properties were produced with the aim of limiting bacteria infections, currently one of the main complications in bone surgeries.

Materials and methods

The non-ionic surfactant Pluronic F127 was selected as structure directing agent and the evaporation induced self-assembly (EISA) process was employed for the synthesis of the Gallium doped mesoporous glasses (Ga-MBG). Pluronic was dissolved in ethanol and nitric acid. Glass precursors were added with a 3h interval between each addition in the following order: TEOS, TEP, calcium and silver nitrate. The dried gels were then calcinated.

Results

The resulting materials were characterized by TEM in order to study their inner microstructure and BET to evaluate the specific surface area and the pore size distribution. Bioactivity tests were performed in simulated body fluid (SBF) to evaluate the MBG ability to form a hydroxycarbonate apatite (HCA) layer on the surface and to confirm that the material can bond to bone. Moreover, the antibacterial capability of Ga-MBGs was confirmed by evaluating Ga-MBGs particles against *Staphilococcus Carnosus* (Gram+) and *Escherichia Coli* (Gram-).

Conclusion

MBGs are optimal candidates to be used as local delivery system of drugs, biomolecules, and therapeutic metallic ions. Thanks to their unique structure they reduce the risk of dangerous side effects to the patients and can confer to the material a tailored controlled delivery of the drug. Ga-doped MBGs are attractive antibacterial systems with potential for bone regeneration applications.

Ackowledgements

This research was carried out with the financial support of the EU Horizon 2020 framework project COACH (ITN-ETN, Grant agreement 642557)

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P086

Regulation of the behavior of HUVEC-cells on polyelectrolyte multilayer coated surfaces

*A. Rudt[1], X. Xiong[2], R. Krastev[1]
[1]Hochschule Reutlingen, Intelligente Oberflächen , Reutlingen, Deutschland
[2]NMI Reutlingen, Biophysikalische Chemie , Reutlingen, Deutschland

Implant-tissue interaction can be directly controlled by surface modifications where the contact interface is precisely tuned according to therapeutic purpose. One of the main challenges of implantation is the undesired excessive acute immune response against the foreign body. Here the transcription factor NF-κB plays a central role in a variety of signaling pathways regulating cell adhesion, proliferation, inflammatory and responses etc.. We are focusing on the understanding of biological response to specific surface properties from an immunological perspective. The goal is the development of a surface coating which can suppress immune response by change of surface properties.

Polyelectrolyte multilayer coatings prepared via layer-by-layer deposition technique enable formation of well-defined surfaces with high variability of the surface properties. In our work we demonstrate a clear correlation between the physical properties of the PEM and NF-κB signaling after the cells were in contact with the PEMs.

The coatings were prepared using different polyelectrolytes. The surface charge, charge density, stiffness and the topography/roughness of the surfaces was varied. Two representative coatings were selected for detailed characterizations. The physical properties of the coatings were characterized by Atomic Force Microscopy (AFM). Tapping mode configuration in liquid environment was used to analyze the interface topography. Force spectroscopy was applied to measure the Young"s Modulus. The immunological reaction was characterized using immunostaining and quantification of activated NF-κB inside the nucleus. Further the downstream produced cytokines and chemokines of the NF-κB pathway were quantified by real time-PCR at transcriptional level.

One coating showed excellent compatibility to HUVEC cells where the cells could well adhere and proliferate. Most interestingly, in this case the NF-κB expression is significantly reduced. On the contrary, other coatings show reduced cell adhesion, metabolic activity and increased immunogenic response.

Based on the data, a correlation between the physical properties of the surfaces and physical strain to the response of the biological cells is demonstrated. This knowledge can contribute to the understanding of such interactions on the interfaces and can guide the further development of biofunctionalized surfaces.

Finacial support: "Innovative Projekte / Kooperationsprojekte an den Hochschulen Baden-Württemberg".

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P087

New bone substitutes for bone defects: Biomechanical analysis in a clinical test set-up of tibial head fractures.

P. Heilig[1], T. Brückner[2], M. C. Jordan[1], M. M. Paul[1], T. Blunk[1], R. H. Meffert[1], U. Gbureck[2], *S. Hoelscher-Doht[1]
[1]University Clinics of Würzburg, Department of Trauma, Hand, Plastic and Reconstructive Surgery , Würzburg, Deutschland
[2] University of Würzburg, Department for Functional Materials in Medicine and Dentistry , Würzburg, Deutschland

Introduction

Bone substitutes are commonly used for filling up bone defects like in tibial head fractures. Here, the biomechanical properties of such materials are of high importance, however, data on this in literature is rare. Thus, this study investigates the basic biomechanical characteristics of different bone substitutes, the screw-bone substitute interface and the combination of substitute and screw osteosynthesis in a biomechanical fracture model for tibial head fractures.

Methods

An experimental drillable apatite cement with poly-2-hydroxyethyl methacrylate-hydrogel additive [1] and experimental magnesium phosphate cement (MPC) were compared with commercially available Graftys® Quickset (apatite). Compressive strength and pull-out strength were determined (Figure 1a). In a tibial head fracture model [2], the bone substitutes were applied for filling up the bone defect alone and in combination with a screw osteosynthesis (Figure 1b). Displacement of the fracture fragment, maximum load and stiffness were calculated in cyclic and maximal axial loading tests (Figure 1c).

Results

The MPC revealed a significant higher compressive strength, pull-out strength and stiffness and a significant lower displacement (Figure 2) compared to the other bone substitutes. For the combination with screws, all bone substitutes revealed higher maximum loads and stiffness values. For the drillable calcium phosphate cement a significant higher displacement (Figure 2) was determined compared to the other bone substitutes.

Conclusions

MPC provides a high biomechanical stability in the pure material testing series and also in the substitute-bone interaction tests. Due to a low viscosity, the cement revealed a high integration in the spongiosa and a complete filling up of the bone defect around the placed screws. For tibial head fractures, only the combination of bone substitute and screw osteosynthesis provides under lower and maximal loading conditions an adequate stability.

References

[1]Christel T. et al., J Mater Sci Mater Med, 2013. 24(3):p. 573-81.
[2]Hoelscher-Doht S. et al., J Orthop Sci, 2014. 19(6):p. 978-83.

Figure 1: The test set-up for the screw pull-out test (a) and the axial loading of the tibial head (c) are shown. The bone substitutes were applied in combination with screws for stabilization of tibial head fractures (b).

Figure 2: The results of the displacement of the fracture fragment of the tibial head are shown. Significant differences are marked by *.

Figure 1

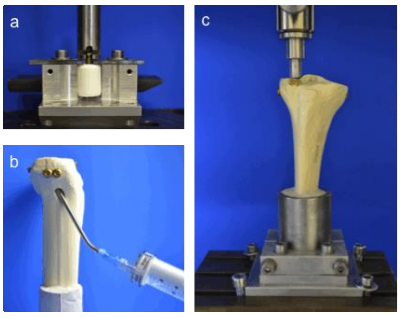
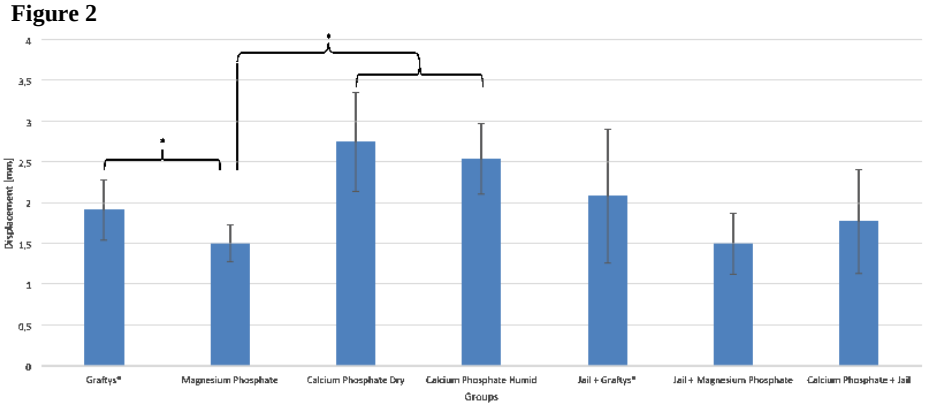


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P088

Evaluation of in-vitro biocompatibility and -cytotoxicity of soft silicone-based polyurea-elastomers for application as accommodating intraocular lens.

*N. Riehle[1,2], F. Klösel[3], S. Thude[4], S. Thanos[3], G. E. M. Tovar[4,5], G. Lorenz[1,2]
[1]Hochschule Reutlingen, Reutlingen Research Institute , Reutlingen, Deutschland
[2]Hochschule Reutlingen, Fakultät Angewandte Chemie , Reutlingen, Deutschland
[3]Universitätsklinikum Münster, Institut für Experimentelle Ophthalmologie , Münster, Deutschland
[4]Fraunhofer Institut für Grenzflächen- und Bioverfahrenstechnik , , Stuttgart, Deutschland
[5]Universität Stuttgart, Institut für Grenzflächenverfahrenstechnik und Plasmatechnologie , Stuttgart, Deutschland

Introduction

Silicone-elastomers found broad applications in medical devices and implants due to their high biocompatibility/-stability, adjustable Young"s Modulus and oxygen permeability. [1,2] Silicone-based polyurethanes were established to combine flexibility, mechanical stability and enhanced biostability for their application in long-term implants. [3]

In this study, selected silicone-based polyurea-elastomers (PU), which were prepared for the application as an accommodating intraocular lens, were tested for their biocompatibility and cytotoxic potential in-vitro.

Methods

In-vitro cytotoxicity tests were performed on HaCaT-cells using an accredited test method according to DIN EN ISO 10993-5:2009. PU-elastomers were extracted with serum-free cell culture medium for 72h ± 2h. After addition of 10% FCS, cells were incubated for 24h ± 2h and cell viability was determined using the water soluble tetrazolium dye MTS. The cytotoxic potential was calculated with the help of controls (negative, positive and blind).

In-vitro biocompatibility was tested using 2 retinal cell-lines: SSW-61 and ARPE-19 cells which are mouse photoreceptor cells and human retinal pigment epithelial cells, respectively. 2x2 mm cut PU-samples were used as substrates for cultivation of mouse and human retinal cells. Cell morphology, -density, proliferation and indications for apoptosis were controlled daily, for 4 days until cells were grown confluent. Additionally, PU-elastomers were co-cultured with 3-dimensional pieces of retinal tissue from rats.

Results and Conclusion

All tested PU-elastomers did not show any cytotoxic effect and were biocompatible with human and mouse retinal cells. Cell proliferation and morphology of SSW-61 and ARPE-19 cells on PU-elastomers and multiwell-plates were equal. No signs of apoptosis within the test-period were detected and PU-materials did not show any deposits or induced adverse effects on layering of the retinal tissue.

Acknowledgments

Financial support of this project by the German Federal Ministry of Education and Research (BMBF) under grant number 03FH03213 is gratefully acknowledged.

References

[1] S. L. Qizhi Chena, George A. Thouas, *Progress in Polymer Science*, 2013, **38**,584-671.
[2] P. Ducheyne, "Comprehensive Biomaterials", Elsevier, 2011.
[3] A. Simmons, A. D. Padsalgikar, L. M. Ferris, L. A Poole-Warren, *Biomaterials*, 2008, **29**, 2987-29995.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P089

Synthesis and Characterization of NIR Dye-Doped Nanoparticles for in vivo Tumor Diagnostics

*C. Schneider[1], S. Dembski[2,3], H. Walles[3,1]
[1]Lehrstuhl Tissue Engineering und Regenerative Medizin (TERM), Universitätsklinikum Würzburg, , Würzburg, Deutschland
[2]Fraunhofer Institut für Silicatforschung ISC, , Würzburg, Deutschland
[3]Translationszentrum Würzburg "Regenerative Therapien für Krebs- und Muskuloskelettale Erkrankungen« TZKME, Zweig des Fraunhofer IGB, , Würzburg, Deutschland

Nanoparticles (NPs) are promising tools for a wide spectrum of biological and medical applications. They can be used as carrier and delivery systems for active agents such as biomolecules, dyes and a wide range of sensitive substances and also contribute to the stabilization of these compounds *in vivo*. Real time non-destructive imaging screening *in vivo* can be performed by means of fluorescent based methods. Near infrared (NIR) dyes are perfectly suited for this purpose. They are very promising for tissue labeling because of the fact that in the IR range there is significantly lower background fluorescence than in the visible range. Another feature of tissue is the so called transparent "NIR-window" at wavelengths from 650 nm to 1350 nm. One major disadvantage of most organic NIR dyes is their very fast degradation *in vivo*, so long-term investigations are not feasible. To stabilize these dyes, one option is to encapsulate the dye molecules into a NP matrix.

Here, we present our recent research activities in the field of medical diagnostics concerning the encapsulation of NIR dyes into NPs for *in vivo* tumor diagnostics. Our work is focused on the synthesis and characterization of NP carrier systems on the basis of amorphous silica with mean particle sizes in the range of 60 to 150 nm. These NPs are synthesized via wet-chemical synthesis and doped with different NIR dyes. The choice of silica as a basis of the NPs is motivated by their high biocompatibility, biodegradability and the possibility of surface modifications.

The characterization of the NPs is done by conventional methods such as transmission electron microscopy (TEM) and dynamic light scattering (DLS). Dye-doped NPs were characterized by fluorescence spectroscopy, measuring of absorption and emission with a plate reader and elemental analysis. The focus here was on the stability of the encapsulated NIR dyes under different storage conditions.

In summary, the synthesis of different NP systems on the basis of amorphous silica and the encapsulation of different NIR dyes was successfully demonstrated. With the confirmation of the stability of the encapsulated dyes in the NP matrix they have shown their potential in the field of medical imaging.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P090

In vitro-Untersuchung neuer Materialien für die additive Herstellung von Epithesen, Septumbuttons und Otoplastiken sowie Gehäuse für Im-Ohr-Hörgeräte

*K. Otto[1], A. Enkelmann[1], G. Grimm[2], R. Wyrwa[2], M. Schnabelrauch[2], G. Schneider[1]
[1]Universitätsklinikum Jena, HNO Forschungslabore/ AG Biomateriallabor , Jena, Deutschland
[2]Innovent e.V. , , Jena, Deutschland

Ziel ist die Entwicklung neuer biokompatibler Materialien und Materialkombinationen für ein additives Herstellungsverfahren sowie einer dehäsiven Oberflächenbeschichtung zur Verringerung der Biofilmbildung. Die zu entwickelnden Materialien und Beschichtungen sollen verschiedene Einsatzgebiete in der HNO-Heilkunde abdecken. Großflächige Weichgewebedefekte im Kopf-/Gesichtsbereich entstehen häufig durch die operative Entfernung eines Tumors. Für die ästhetische Rekonstruktion des Gesichts werden künstliche Gesichtsteile (Epithesen) individuell gefertigt und angepasst. Ein weiteres häufiges Krankheitsbild in der HNO-Heilkunde sind Defekte des knorpeligen Nasenseptums. Diese werden durch individuell gefertigte Prothesen aus Silikon (Septumbuttons) verschlossen. Im Bereich der Hörgerätsysteme werden Ohrpassstücke (Otoplastiken) als Teil der Hinter-dem-Ohr-Hörsysteme sowie Im-Ohr-Hörgeräte, die komplett im Ohr bzw. Gehörgang getragen werden, individuell angepasst, um Halt, Belüftung und bequemen Sitz zu gewährleisten. Das neu entwickelte Elastomer auf Methacrylat-Basis sowie verschiedene Oberflächenbeschichtungen wurden chemisch und thermisch nachbehandelt und anschließend auf Zytokompatibilität geprüft. Dazu wurden Viabilitätsassays (WST-1, LDH) mit etablierten Zelllinien (HeLa, MC3T3-E1) durchgeführt. Die Testung erfolgte sowohl durch direkte Besiedlung der Prüfkörper als auch durch Kultivierung der Zellen mit Eluaten über einen Zeitraum von 24h/72h. Die Besiedlung der Probekörper wurde mittels Methylenblau-Färbung untersucht. Die Kultivierung der Zellen mit Eluaten sowie die direkte Zugabe der Probekörper zu den Zellen führen zu einer hohen Viabilität, vergleichbar mit den Kontrollen. Es zeigen sich keine Unterschiede im Vergleich der chemischen Nachbehandlungen. Eine Hitzebehandlung der Probekörper führte in der Zellkultur zu einer höheren Viabilität. Die Methylenblau-Färbung zeigte keine direkte Besiedlung der Probekörper. Die Ergebnisse zeigen, dass das Methacrylat-basierte Material wärmebehandelt werden muss, da so die Menge an Monomeren mit Methacrylatgruppen minimiert wird. Eine definierte Waschprozedur führt zur weiteren Verringerung toxischer Bestandteile, wodurch eine hohe Zytokompatibilität erreicht wird. Ein unerwünschtes Zellwachstum fand auf den untersuchten Oberflächen nicht statt. Wir konnten zeigen, dass das entwickelte Material oberflächenbehandelt werden kann und für die oben genannten Einsatzgebiete sehr gut geeignet ist.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P091

Histological and histomorphometrical analysis of an individually fitted allogenic bone block for ridge augmentation

O. Blume[1], *P. Donkiewicz[2], B. Buchbinder[2], M. Back[1], S. Wenisch[3], R. Schnettler[4], M. Barbeck[5,6]
[1]Praxis im Tal, , München, Deutschland
[2]botiss biomaterials GmbH, , Zossen, Deutschland
[3]Klinik für Kleintiere, Institut für Veterinär-Anatomie, -Histologie und –Embryologie , Gießen, Deutschland
[4]Universitätsklinikum Gießen, Klinik und Poliklinik für Unfallchirurgie , Gießen, Deutschland
[5]botiss biomaterials GmbH, Research & Development , Zossen, Deutschland
[6]Charité - Universitätsmedizin Berlin, Julius Wolff Institute and Center for Musculoskeletal Surgery, Berlin-Brandenburg Center for Regenerative Therapies , Berlin, Deutschland

Objective

To date the treatment of alveolar bone defects especially within the esthetic zone, with respect to both functional and esthetic restoration, remains challenging. While the clinician"s options for handling of such defects used to be limited to the utilization of autologous bone grafts (ABG), the advancing development of bone substitute materials created a set of alternatives with which comparably predictable clinical outcomes can be achieved. As they inherit essential biological and structural features of ABG, freeze-dried bone allografts (FDBA) represent the most promising option for healing of bone defects. The present case report demonstrates complex bone regeneration of a 43 year old woman who presented with periimplantitis related to three dental implants which led to massive bone resorption and partial loss of the buccal wall within the esthetic zone.

Material and Methods

Following implant extraction, an individually fitted FDBA block (maxgraft® bonebuilder, botiss biomaterials GmbH, Berlin) and a resorbable collagen membrane (Jason® membrane) were used for GBR surgical procedure. At re-entry, six months after surgery, biopsies were taken and implants were placed.

Results

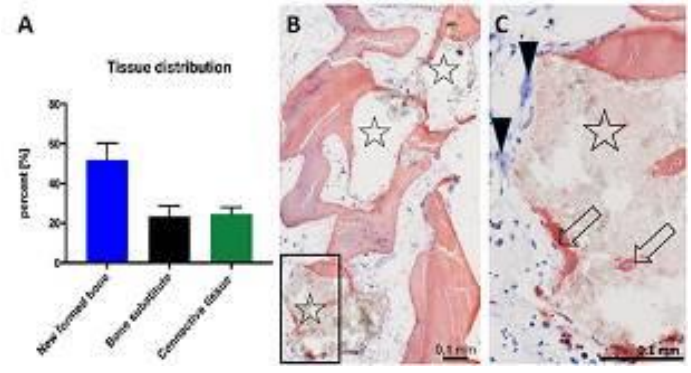
Stable and vital new formed bone was found in the augmentation area at re-entry resulting from the FDBA material (Fig. 1 B). No histological signs of implant-related inflammatory tissue reactions were observed (Fig. 1 C). The healing course was uneventful resulting in minimal material resorption. Histomorphometrical analysis verified the remodeling capacities of FDBA resembled by the formation of new bone (52 %), which was evidently exceeding the amounts of connective tissue (25 %) and residual grafting material (23 %) respectively (Fig. 1 A). X-ray scans taken 6 and 9 months post-OP indicated complete remodeling of the allogenic bone block, almost no resorption and hence stable implantation into the new formed bone providing an optimal result.

Conclusion

The presented case demonstrates the excellent clinical performance of a customized bone block based on FDBA to be a reliable alternative to ABG even in spacious and complex alveolar ridge defects.

Fig. 1: Results of the histomorphometrical analysis at six months post-OP (**A**) and exemplary histological images (**B and C**). Residual biomaterial (asterisks), new formed bone (arrows), multinucleated giant cells (arrowheads).

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P092

Expression of modified HGF for the tunable release from high performance ceramic implant materials

*S. Reinhold[1], F. Böke[2], N. Labude[1], *L. Zenner[1], C. Schmitz[3], I. Lauria[2], W. Jahnen-Dechent[3], H. Fischer[2], S. Neuss[1,3]
[1]RWTH Aachen University, Institute for Pathology , Aachen, Deutschland
[2]RWTH Aachen University, Dental Materials and Biomaterials Research , Aachen, Deutschland
[3]RWTH Aachen University, Helmholtz Institute for Biomedical Engineering, Biointerface Group , Aachen, Deutschland

High performance oxide ceramics like alumina and zirconia are versatile biomaterials used primarily for articulating components of artificial joints. Although superior to the current gold standard titanium in various regards, their major drawback is a lack of osseointegration *in vivo*, resulting in fibrous encapsulation and ultimately loss of implant stability.To overcome this limitation both tissue integration and recruitment of stem cells towards the implant have to be enhanced. For the latter, hepatocyte growth factor (HGF) is a key factor for mesenchymal stem cell (MSC) recruitment.The aim of this project is to modify wild-type HGF to contain a highly specific biomaterial coupling site combined with an enzymatic tPA cleavage sequence for the tunable release of HGF during wound healing situations *in vivo.* To this end, a cysteine tag was introduced at the N-terminus for immobilization on ceramic surfaces. This is followed by a spacer chain to allow for plain enzyme access. At first a proprietary HGF sequence from Spintec GmbH containing an N-terminal tPA-cleavage site and spacer molecule was extended by a single cysteine via PCR using a mutagenic primer pair. The resulting template was cloned into a TOPO TA vector and used to transform *E. coli Top10* cells. Sanger sequencing validated correct vector assembly. Trex FlpIn cells were then co-transfected with this vector and a helper plasmid for a stable genomic integration. Clones were selected via antibiotic resistance towards hygromycin B. Successful genomic implementation was confirmed via PCR, Sanger sequencing and zeocin sensitivity. Following cell culture incubation, HGF secretion into the medium supernatant was significantly higher for the transfected cells, as shown by western blot analyses. Protein purification was achieved by affinity chromatography). Functionality of the modified HGF was proven in Boyden chamber assays. Secreted HGF after modification showed nearly the same affinity to induce motility in HuH7 cells in comparison to 75 ng/mL recHGF (Sigma). This suggests that we generated a stable cell line that is able to express a functional modified HGF variant. Preliminary functional assays indicated HuH7 migration towards a modHGF gradient. Our findings thus demonstrate the chemoattractant properties of the newly expressed modHGF. In conclusion, the demonstrated HGF recombination presented here leads to the expression of functional proteins for further studies regarding tailored MSC attraction.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P093

Antimicrobial and Cytotoxicity Evaluations of Hydrogen peroxide- Towards Clinical Application of Antimicrobial Biomaterials for Wound Dressings

*A. Idrees[1,2,3], S. Pacharra[3], L. Marlinghaus [4], V. Chiono[1], G. Ciardelli[1], J. M. Vasquez[1], U. Greiser[5], W. Wang[5], R. Viebahn[2], S. G. Gatermann[4], J. Salber[2,3]
[1]Politecnico di Torino, Department of Mechanical and Aerospace Engineering , Torino, Italien
[2]Knappschaftskrankenhaus Bochum, Hospital of RUHR-University Bochum, Clinic of Surgery , Bochum, Deutschland
[3]RUHR University Bochum, Clinical Research Center , Bochum, Deutschland
[4]Ruhr-University Bochum , Department of Medical Microbiology , Bochum, Deutschland
[5]Vornia Biomaterials Ltd, , Dublin, Irland

Chronic wound infections and emerging drug resistance are serious problems causing a high morbidity and cost of healthcare. Therefore, investigation on novel antimicrobial strategies is of great interest. Use of honey from ancient times is reputed for its wound-healing and antibacterial properties. Along with other factors, major antibacterial factor in honey is hydrogen peroxide (H2O2), which is produced by Glucose Oxidase (GO). This approach can be employed for polymer based novel antimicrobial biomaterials for wound dressings.

In this study, the inhibitory effect of H2O2 on numerous bacteria of clinical significance, was investigated. To determine the "safe" antimicrobial concentration of H2O2, cytocompatibility analysis was also performed for cellular cytotoxicity. The effect of externally added H2O2 was performed by exposing L929 fibroblasts to various H2O2 concentrations. At different time points, cell viability was assessed by a measure of cell metabolic activity, cell membrane integrity and cell morphology. Whereas, antimicrobial efficacy was evaluated against a wide range of pathogenic bacteria involved in chronic wounds namely S. aureus, S. epidermidis, S. lugdunensis, E. faecalis, E. coli, P. aeruginosa, K. pneumoniae, and A. baumanii. Antimicrobial tests were performed using broth microdilution method for the determination of Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC).

Results showed different MIC and MBC values for different bacterial species indicating their differences in susceptibility to treatment. Among the tested bacteria, S. aureus showed 99.9% bacterial reduction at the concentration of 0.5mM, while E. faecalis showed the highest MIC value of 5mM. A. baumanii is one of the most pathogenic bacteria involved in serious skin wound infections also showed the same MIC value (0.5 mM) as S.aureus. Cytotoxicity results showed two distinct patterns in our experimental set-up: the highest concentrations rapidly induced cell death characterized by morphological evidence and plasma membrane damages as compare to the concentrations of 1 mM and 100 µM where the cytotoxic effect only increased gradually over time indicating concentration dependent distinct pathways of H2O2-induced cytotoxicity. Cytotoxic effects would differ depending on "at once" or "gradual" (as can be produced by entrapped GO and glucose) H2O2 exposure determining the ability of cells to detoxify H2O2 that needs further investigation.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P095

Expansion and myogenic differentiation of adipose-tissue derived mesenchymal stem cells for application in a cell-seeded vessel graft

*K. Müller[1], M. Bienert[1], N. Labude[1], S. Neuß[2]
[1]Uniklinik RWTH Aachen, Institut für Pathologie , Aachen, Deutschland
[2]Uniklinik RWTH Aachen, Institut für Pathologie und Helmholtz Institut für biomedizinische Technologien - Gruppe für Zell- und Molekularbiologie an Grenzflächen , Aachen, Deutschland

The increasingly elderly population of Germany suffers more and more from limitations like cardiovascular diseases, diabetes mellitus or impaired renal functions (Bundesamt für Statistik, 2013; Bundesamt für Statistik, 2016a; Bundesamt für Statistik, 2016b).

The treatment of these diseases can benefit from the development of an arterial vessel implant with a small diameter (<5mm) (Canver, 1995). For avoiding complications like stenosis or aneurysms and for offering the best compatibility at the same time, this implant should imitate the natural layers of an arterial blood vessel (Kawamoto *et al.*, 2015). Especially the *media* consisting of smooth muscle cells (SMC) is focused. SMC can be differentiated from AD-MSC (adipose-tissue derived mesenchymal stem cells), that can be isolated easily autologously and in big amounts. The StemGraft was already applied successfully in sheep and is now facing the adaptation for the needs of the clinical application in humans (Koch *et al.*, 2010).

For the development of this stent AD-MSC are expanded and myogenically differentiated in xenofree and thus GMP-compliant media. Both AD- and BM-MSC (bone marrow-derived mesenchymal stem cells) grow with reduced cell size, but fibroblastic morphology in xenofree culture. Cell counting during the culture shows, that the proliferation of both cell types is highly increased compared to FCS containing conditions and the stem cell phenotype remains stable. The cell number needed for seeding a StemGraft is 100 million of cells and is reached after at least three passages in xenofree culture. The culture expansion time can possibly be shortened or even be completely circumvented by isolating more AD-MSC out of more extracted tissue.

Cells being cultured in xenofree media produce an extracellular matrix. Immunohistochemical stainings show the content of proteins of the natural extracellular matrix, that possibly supports cell growth.

A significant increase of the smooth muscle cell protein expression analysed by flow cytometry compared to the expansion control can only be detected in differentiation with TGF-β1 and BMP4 in FCS containing conditions. However, the basal expression of these markers in expansion conditions is already comparable to that of the positive control, so even undifferentiated cells can be used.

In summary the preparation of the cellular components of the small-diameter, cell-seedes vessel graft for the adaption to human application was successful.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P096

Application of magnetic cell seeding technique for improved cell colonization of biomaterials

*R. Singh[1], A. Wieser[1], S. Reakasame[2], B. Dietel[3], R. Detsch[2], A. R. Boccaccini[2], C. Alexiou[1], I. Cicha[1]
[1]University Hospital Erlangen, Section of Experimental Oncology and Nanomedicine (SEON), Cardiovascular Nanomedicine Unit, Department of Otorhinolaryngology, Head and Neck Surgery , Erlangen, Deutschland
[2]Institute of Biomaterials, University of Erlangen-Nuremberg , Erlangen, Deutschland
[3]University Hospital Erlangen, Med 2, Department of Cardiology and Angiology , Erlangen, Deutschland

Introduction

Poor initial attachment of cells and non-uniform colonization may contribute to the failure of tissue-engineered scaffolds. Cellular adhesion to matrices can be enhanced by using cells loaded with superparamagnetic iron oxide nanoparticles (SPIONs) and external magnetic force. We tested the suitability of magnetic seeding technique for improving the attachment and growth of fibroblasts and endothelial cells on different alginate-based hydrogels.

Materials and Methods

Hydrogels containing alginate (Alg), alginate dialdehyde crosslinked with gelatin (ADA-G) and Alg blended with G or silk fibroin (SF) were placed in 24-well plates. Endothelial cells (ECs) and fibroblasts were loaded with SPIONs (ECs: 3 µg/cm2; fibroblasts: 5 µg/cm2) for 24h, followed by seeding and culture on hydrogels for up to 7 days, in the presence or absence of magnetic field during the first 24h. Cell morphology (F-actin), viability (calcein) and metabolic activity (WST-8 assay) of magnetically-seeded versus conventionally seeded cells were compared.

Results

Compared with conventional method, magnetic seeding dramatically enhanced the initial fibroblast coverage on ADA-G and on Alg/SF, leading to the formation of a dense cellular layer already at day 1 and axial orientation from day 3 on. Magnetic seeding of ECs improved their initial attachment to Alg/G hydrogels. After 7 days of culture, the difference in metabolic activity between magnetically and conventionally seeded ECs became less distinct, but a more pronounced monolayer formation and cobblestone morphology were observed in magnetically-seeded samples. In contrast, we did not achieve an efficient and stable colonization of ADA-G films with ECs even with magnetic seeding. On pure Alg, initial attachment of magnetically-seeded cells was dramatically improved compared to the conventional method, but this effect was transient and diminished with the cessation of magnetic force. A gradual rounding and clustering of cells on Alg was observed after day 3, indicative of reduced cell-material contacts.

Conclusion

Magnetic cell seeding improves the initial cell attachment to hydrogel surface, which can shorten culture time and may thus play a decisive role for the regenerative outcomes. However, on less cell-compatible hydrogels, the long-term efficacy of cell colonization after the cessation of magnetic field is primarily dependent on the material properties that govern the subsequent cellular responses.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P097

Manganese containing mesoporous bioactive glass nanoparticles as a drug delivery system: fabrication and biocompatibility

*Q. Nawaz[1], M. A. Ur Rehman[1], A. R. Boccaccini[1]
[1]Institute of Biomaterials, Department of Material Science and Engineering, University of Erlangen-Nuremberg , Erlangen, Deutschland

Multifunctional mesoporous bioactive glasses incorporating different biological active metallic ions can be also designed to deliver therapeutic drugs. This has attracted the attention of researchers working on bone tissue engineering and wound healing. Manganese (Mn) is a therapeutically beneficial ion, which has potential to improve osteogenic differentiation and cell adhesion. In this study, a mesoporous bioactive glass particles based on the system: SiO2–P2O5–CaO–MnO was developed via a modified Stöber process. Natural herb Icariin (potential osteoinductive compound) was loaded into synthesized Mn-MBG particles. Mn-MBG was characterized by Scanning electron microscopy (SEM), Energy dispersive X-ray spectroscopy (EDX), X-ray diffraction (XRD), Fourier transform infrared spectroscopy (FTIR), Raman spectroscopy, Brunauer–Emmett–Teller (BET) and Ultraviolet visible spectroscopy (UV-VIS). Results indicate that the Mn-MBG particles have following properties: spherical morphology of mean diameter 130 nm, amorphous structure, average pore size of 7.65 nm and specific surface area of 3.697 x 102 m2/g. Furthermore, upon immersion in simulated body fluid (SBF), a layer of carbonated hydroxyapatite (cHA) formed on pellet made from the Mn-MBG powder confirming the bioactivity of material. The previous results indicate that Mn incorporation into the bioactive glass network is an effective strategy to develop novel bioactive glasses for dual drug and therapeutic ion release for bone tissue engineering.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P098

Increasing biocompatibility of patient specific PEO-coated magnesium implants using endothelial cells and mesenchymal stem cells in bone defects

*M. Bienert[1], L. Jauer[2], M. Muether[3], C. Ptock[3], B. Lethaus[4], S. Neuss[1]
[1]RWTH Aachen University, Helmholtz Institute for Biomedical Engineering - Biointerface Laboratory and Institute of Pathology , Aachen, Deutschland
[2]RWTH Aachen University, Fraunhofer Institute for Laser Technology ILT , Aachen, Deutschland
[3]Meotec GmbH & Co. KG Aachen, , Aachen, Deutschland
[4]RWTH Aachen University, Department of Oral and Maxillofacial Surgery University Hospital , Aachen, Deutschland

Bone graft vascularization is a main challenge in tissue engineering to improve biocompatibility and clinical translation of bone constructs. Osseointegration and reconstruction of function after implantation can be achieved by support of a bone environment rich in vascular networks. In here structural and vascular integration is achieved by polyethylene oxide (PEO)-coated magnesium grafts cultured with autologous mesenchymal stem cells (MSC) and endothelial progenitor cells from peripheral blood (EPC) to increase biocompatibility. Magnesium grafts for craniofacial defects are designed in a patient-specific way using DICOM data from the Department of Oral and Maxillofacial Surgery University Hospital Aachen.

Materials are bated before PEO-coating by the company Meotec Aachen and coated and non-coated grafts are compared. To enable EPC and MSC culture on the materials, materials need to be sterile before they are used in cell culture. In figure 1, four different prevalent sterilization methods are tested. Since none of the sterilization methods changes the topography of the grafts we decided to use UV-C light exposure to sterilization the materials for cytotoxicity testing. Compared to the other methods UV-C provides a gently method for sterilization.

Cytotoxcity studies for different magnesium grafts are performed according to ISO 10993-5. Magnesium grafts with or without PEO-coating are incubated for 24h at 37°C in cell culture medium. Human umbilical vein endothelial cells (HUVEC), human MSC or L929 cells are seeded on cell culture plastic and are allowed to adhere for one day. Media incubated with grafts is added to respective cells and incubated for additional 24h. Afterwards cells are stained with fluorescein diacetate (FDA) and propidium iodide (PI) and evaluated with fluorescence microscopy. Figure 2 shows the live/dead staining of the respective materials where viable cells are green fluorescent and necrotic cells are red fluorescent. Bating of the magnesium grafts significantly reduces HUVEC viability compared to negative control and PEO-coated material. This indicates that PEO-coating is essential for HUVEC viability. Neither bating nor bating with subsequent PEO-coating of the magnesium grafts showed a cytotoxic effect on MSC or L929 cells.

Figure 1

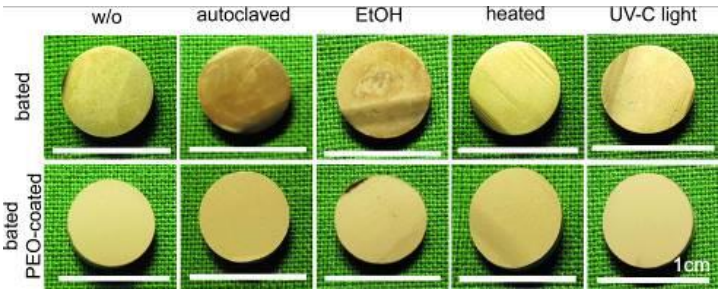
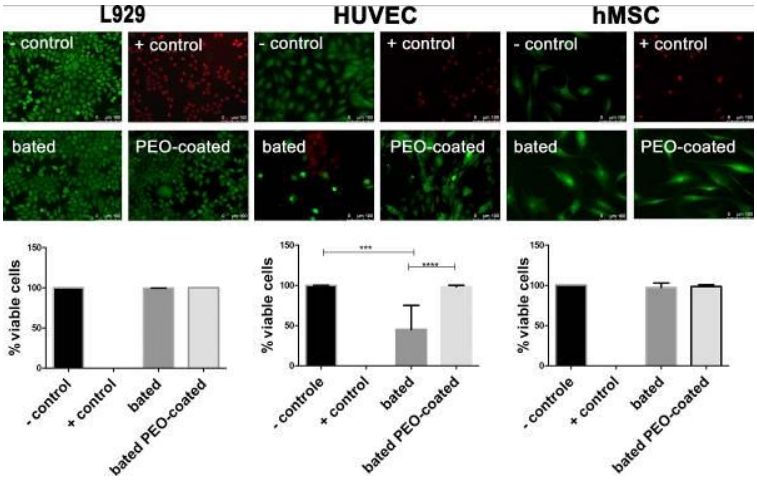


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P099

Obtaining materials based on cryogel of polyvinyl alcohol and gelatine for formation of artificial blood vessels

*A. Plyaskina[1], D. Lytkina[1], I. Kurzina[1]
[1]National Reseach Tomsk State University, , Tomsk, Russische Föderation

Cardiovascular diseases (CVD), in particular atherosclerosis, is one of the most socially significant diseases. To eliminate these diseases, vascular grafts are used, which should have such important properties as biocompatibility, porosity and elasticity [1]. One of the most promising materials are materials based on polyvinyl alcohol cryogels (PVA), since they have all the required properties and are biodegradable. The aim of this work was to obtain the biocompatible material based on PVA cryogel, which will similar the mechanical and hydrodynamic properties of the average vessel of the man. Synthesis of the material was carried out according to the following scheme: gelatine powders were added in different ratios to the 8% solution of PVA (At a molecular weight of 85,000-124,000 and at 99 +% hydrolyzed) (Table 1). The prepared mixtures were poured into a mold to produce tubes with an internal diameter of 3 mm. Further, the formed tubes were subjected to several cycles of freezing / thawing at -80 °C. The composites were then kept in water at 40 ° C for 72 hours to remove gelatin [2]. Infrared spectra indicate that the resulting materials are a porous PVG cryogel.

Table 1. The composition and porosity of the samples

Analytical band amide (1650-1680 cm-1) corresponding to gelatin is completely absent. Figure 1a shows micrographs of the surface of samples and cryogel PVA, the dimensions and distribution of pores in Figure 1b were calculated from them. The diagram shows that with increasing gelatin concentration in the samples, the pore sizes and their ratio increase. Porosity distribution is monomodal. Calculation of the porosity of the materials with respect to cryogel PVA was carried out by gravimetric method [3].

Figure 1. a) Microphotographs of the surface of materials and pure cryogel PVA, b) Pore distribution size

By the results of the experiment, it is evident that an increase in the gelatin concentration leads to a sharp increase in porosity to 60% (Table 1). As a result of the work done, porous tubular materials based on PVA cryogels were obtained, the pore size and porosity of which can be controlled by the amount of gelatin added.

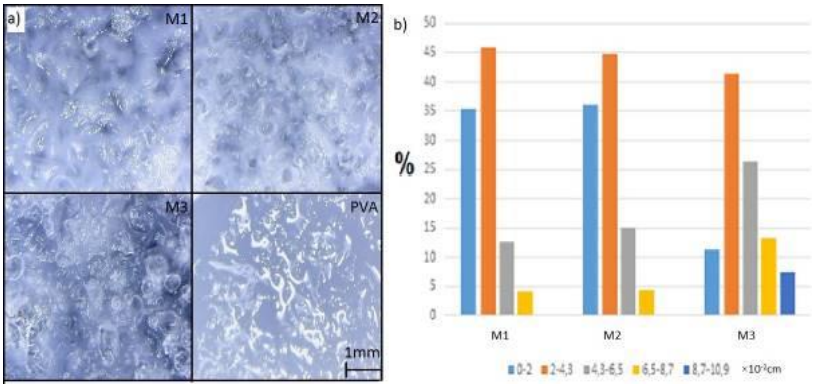
References

[1] Teresa Simon-Yarza., et al. Journal of Cardiovascular Translational Research, 2017, 1-14.
[2] Jia-Horng Lin et al. Journal Materials Letters, 2015, 190, 201–204.
[3] GOST ISO 7198-2013. Cardiovascular implants - Tubular vascular prostheses.

Figure 1

Samples	M1	M2	M3
Concentration of gelatin, g / 10 ml	0,4	0,8	1,2
Porosity , %	4,3	31,5	59,3

Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P100

Electrospun Zein Based Scaffolds for Cardiac Tissue Engineering

*L. Vogt[1], L. Liverani[1], A. R. Boccaccini[1]
[1]Friedrich-Alexander-Universität Erlangen-Nürnberg, Lehrstuhl Biomaterialien , Erlangen, Deutschland

Introduction

Due to the very limited intrinsic regeneration ability of cardiac tissue after an event like myocardial infarction, strategies to repair the damaged tissue are of particular interest [1]. To obtain a functional scaffold for cardiac tissue engineering (CTE), the technique of electrospinning has gained much attention recently, since it allows to closely mimic the highly branched myocardial structure and its properties [2]. As materials for biomedical applications, plant proteins are becoming increasingly attractive. Zein, a class of prolamine protein found in corn, is biocompatible, antimicrobial and has low toxicity, but lacks good mechanical properties and hydrolytic stability [3, 4].

Objectives

This study focused on the development of an electrospun scaffold based on the natural protein zein for CTE. To overcome the shortcomings of zein, it was blended with poly(glycerol sebacate) (PGS). PGS is interesting for CTE due to its biocompatibility and tunable mechanical and degradation properties [2].

Materials & Methods

Neat zein and zein blended with PGS prepolymer or mildly crosslinked PGS in different ratios were electrospun. Benign solvent, like acetic acid, was used to enhance the scaffold biocompatibility. Morphological, chemical and mechanical properties and degradation behavior in PBS of the obtained scaffolds were investigated.

Results

Neat zein and zein/PGS with high zein content fiber mats showed defect-free microstructures. The addition of PGS decreased the average fiber diameter, which varies from 700nm to 90nm, depending on the PGS content. It was also found that increased stirring time in acetic acid led to short strand zein fibers. The ultimate tensile stress and failure strain increased with the addition of PGS. Degradation tests showed the morphological instability of zein containing fibers in contact with aqueous media, which caused the collapse of the fibrillary structure.

Conclusion

The fabrication of zein and zein/PGS electrospun scaffolds was successful and revealed promising results for CTE. Short strand zein fibers could find use also as drug carriers. However, to facilitate the use of the fiber mats in aqueous environments, further crosslinking is necessary.

References

[1] J. R. Venugopal, et al., *J R Soc Interface* 2012, 9, 66, 1-19.
[2] R. Rai, et al., *Adv Healthc Mater* 2015, 4, 13, 2012-2025.
[3] D. Dippold, et al., *Eur Polym J 2016*, 75, 504-513.
[4] M. Demir, et al., J *Biomed Mater Res A* 2017, 105.6, 1656-1665.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P101

Development of a degradable dual setting system – combination of PEG-based hydrogels with brushite cement for biomedical applications

*M. Rödel[1], J. Teßmar[1], U. Gbureck[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, Abteilung für Funktionswerkstoffe der Medizin und der Zahnheilkunde , Würzburg, Deutschland

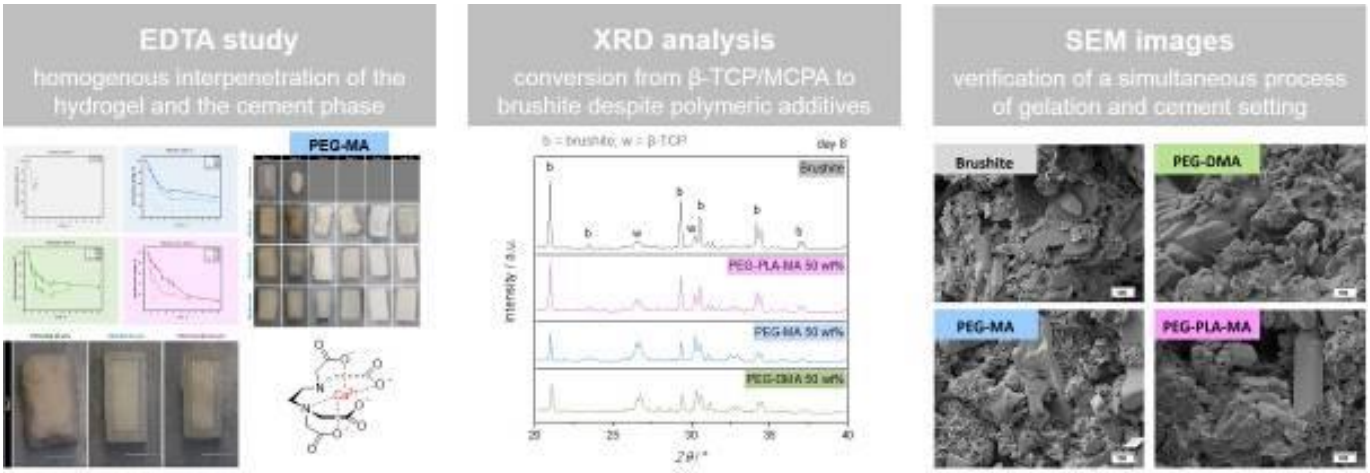
Bone is a hard and elastic tissue which combines an organic component (~ 20 %, mainly collagen type I, responsible for elasticity) with mineral (~ 70 %, mainly hydroxyapatite nanocrystals, responsible for hardness) and water (~ 10 %). [1, 2] With respect to this natural composition, we established a degradable dual setting system of different poly(ethylene glycol) [PEG]-based hydrogels combined with a brushite cement. The idea was to reinforce an inorganic calcium phosphate mineral with an organic phase. In this study, we investigated the phase structure of the composite materials regarding distribution and coherence of the hydrogel and cement via an EDTA dissolution-study, XRD-analysis and SEM-images.

Composite cements were produced by dissolving the hydrogel precursors (low molecular dimethacrylated PEG (PEG-DMA; ~ 550 Da), high molecular PEG-DMA (PEG-MA; ~ 6000 Da) and a variant with additional poly(lactic acid) spacers (PEG-PLA-MA; ~ 6500 Da)) at different amounts in 0.5 M citric acid. A 5 wt% H2O2-solution (radical catalyst) was added to the cement liquid, which was mixed with the cement raw powder consisting of β-tricalcium phosphate (β-TCP) and anhydrous monocalcium phosphate (MCPA) (equimolar ratio) with the addition of 5 wt% ascorbic acid (radical initiator). After homogenous mixing, both the cement setting reaction and the chemical gelation started simultaneously.

Using the decalcifying agent EDTA, the Ca2+-ions of the inorganic phase in the composites were complexed and the whole inorganic phase was dissolved. A higher polymeric content resulted in a more stable remaining hydrogel in the EDTA-solution (10 wt% < 25 wt% < 50 wt%). The analysis of the crack surface via SEM-analysis showed a homogenous incorporation of the hydrogel in the cement phase without any discontinuities or agglomerations. Additional XRD-measurements proved a significant influence of the polymeric matrix on the conversion from β-TCP/MCPA to brushite with a decrease in signal intensity.

The results confirmed a simultaneous process of setting reaction and gelation without an inhibition of the conversion to brushite and the formation of interpenetrating networks of hydrogel and cement.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P102

Studies on osteogenic differentiation of cells on multilayers loaded with BMP-2

*R. Anouz[1], E. Schwarz[2], T. Groth[3,1]
[1]Martin Luther University (MLU), Biomedical Materials Group , Halle (Saale), Deutschland
[2]Martin Luther University (MLU), Institute of Biochemistry and Biotechnology , Halle (Saale), Deutschland
[3]Martin Luther University (MLU), Interdisciplinary Center for Materials Science , Halle (Saale), Deutschland

Introduction

Layer-by-Layer (LbL) is a technique that permits the formation of nanostructured surface coatings useful for delivery of bioactive drugs and proteins, like growth factors (GF). Bone morphogenetic protein-2 (BMP-2) promotes osteogenic differentiation of mesenchymal and other cells and is used in the treatment of non-healing bone fractures.However,bolus injections of BMP-2 are rapidly cleared off and lose their activities upon exposure to blood. Hence, high dosages are needed,which can result in inflammation, increased cancer incidence and high costs. Thus, the need for controlled release systems of GFs are badly needed in the field of regenerative medicine.

Objective

This study was aimed to fabricate various LbL systems using different glycosaminoglycans (GAGs) with capability to bind BMP-2 specifically in order to control osteogenic differentiation of cells by biocompatible release systems. Interestingly is the use of oxidized GAGs for intrinsic cross-linking to improve multilayer stability and also affect the release of GF.

Materials and methods

Heparin, chondroitin sulfate and their oxidized forms as polyanions were combined with chitosan and collagen I as polycations to form various multilayer coatings on model materials. The myoblast cell line C2C12, which can differentiate into osteoblasts was seeded on 5µg/mL BMP-2 loaded multilayers. Cell viability was investigated by Qblue assay; adhesion studies using immunohistochemical staining and osteogenic differentiation by alkaline phosphatase (ALP) assay, ALP and alizarin red-S staining.

Results

C2C12 cells cultured directly on the top of multilayers showed that particularly BMP-2 loaded multilayers made of oxidized GAGs promoted an osteogenic differentiation of C2C12 cells that is nearly comparable to the positive control, when 5µg/mL BMP-2 was added directly to the medium. Interestingly,the BMP-2 had synergistic effect on cell adhesion and spreading; and the loaded BMP-2 affected cell differentiation more than the soluble BMP-2.

Conclusion

The results show that oxidized GAGs forming intrinsically cross-linked mutlilayers are useful as reservoirs for sustained release of BMP-2, which can pave the way for coating implants and scaffolds for repair and regeneration of bone fractures.

Acknowledgments

This work was supported by Bundesland Sachsen-Anhalt in the frame of Leistungszentrum Bio-und Systemtechnik and the International Graduate School AGRIPOL.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P103

Electrospinning of Elastin-Containing Nanofibers

*T. Hedtke[1], T. Kürbitz[2], A. Desai[1,2], C. Schmelzer[1]
[1]Fraunhofer-Institut für Mikrostruktur von Werkstoffen und Systemen IMWS, Biologische und Makromolekulare Materialien , Halle (Saale), Deutschland
[2]Hochschule Anhalt, , Köthen, Deutschland

The extracellular matrix consist of a fibrous network of structural proteins that contributes to the mechanical properties of tissues and provides a scaffold for cell adhesion. Supplying wounds with nanofiber scaffolds supports the reconstitution of the tissue structure and improves the overall process of healing (for review see (1,2)). Therefore the fabrication of protein-based nanofiber materials is a promising method for the development of novel biocompatible and absorbable wound dressings. An established method for producing nanofibers from proteins is electrospinning, albeit the process parameters and solvents need to be determined for each protein or protein mixture.

We are presenting electrospun protein-based nanofiber materials comprising the structural extracellular matrix protein elastin and methods for the production of those. Elastin was purified from bovine aortic tissue by the method of Schmelzer et al. (3) and subsequently partially hydrolyzed by oxalic acid or potassium hydroxide to gain α- or κ-elastin, respectively. The soluble elastin hydrolyzates are utilizable for electrospinning in contrast to the insoluble isolated elastin. α- and κ-elastin were blended with silk fibroin or collagen in different concentrations and process parameters were optimized to reduce bead formation. Different methods for stabilizing the fiber network by covalent cross-linking were tested and the morphology of the fiber mats were characterized by electron microscopy. Fiber form, diameter and the mesh size of the non-woven materials were compared before and after cross-linking. Moreover, the influence of different elastin percentages on the properties of the materials was determined.

The materials produced by the presented methods are potentially applicable as wound dressings, implant coatings or scaffolds for tissue engineering.

References

[1] Chen, S., Liu, B., Carlson, M. A., Gombart, A. F., Reilly, D. A., and Xie, J. (2017) Recent advances in electrospun nanofibers for wound healing. *Nanomedicine* **12**, 1335-1352
[2] Abrigo, M., McArthur, S. L., and Kingshott, P. (2014) Electrospun nanofibers as dressings for chronic wound care: advances, challenges, and future prospects. *Macromolecular bioscience* **14**, 772-792
[3] Schmelzer, C. E., Jung, M. C., Wohlrab, J., Neubert, R. H., and Heinz, A. (2012) Does human leukocyte elastase degrade intact skin elastin? *The FEBS journal* **279**, 4191-4200

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P104

Polydopamine-based copolymerized coating of PCL fiber mats for tissue engineering

*S. Oehmichen[1], A. L. Hoheisel[2], Y. Roger[3], S. Schulze[4], B. Glasmacher[2], A. Hoffmann[3], H. Menzel[1]
[1]TU Braunschweig, Institute of Technical Chemistry , Braunschweig, Deutschland
[2]Leibniz Universität Hannover, Institute for Multiphase Processes , Hannover, Deutschland
[3]MH Hannover, Laboratory for Biomechanics and Biomaterials , Hannover, Deutschland
[4]TU Braunschweig, Institute for Chem. and Therm. Process Engineering , Braunschweig, Deutschland

In times of an aging population, the importance of regenerative therapies, such as tissue engineering (TE), is increasing. In the case of TE, scaffolds are required which enable the bioinspired replication of tissues and the ingrowth of cells. For this purpose, the use of electrospun fiber mats is possible. As a promising scaffold material polycaprolactone (PCL) is often used. It is biocompatible, slowly biodegradable and has good mechanical properties. However, PCL scaffolds have a highly hydrophobic surface, which is unfavorable for cell adhesion. Accordingly, a modification of the surface is necessary. For this a number of methods are known, e.g. plasma treatment [1] or aminolysis [2]. Both methods are able to increase the hydrophilicity, but because they directly attack the structure of the polyester they also affect the mechanical properties. [1,2] Therefore, the interest in thin polymeric coatings has been increased in the last years. A well-known system is the coating with polydopamine obtained by a self-polymerization of dopamine (DA) directly on the surface. [3] The copolymerization of dopamine and hexamethylendiamine (HMD) in a one-step reaction is also known and increases the number of NH2-groups. [4] NH2-functionalities have a high biocompatibility and they are enable subsequent modification with biomolecules containing carboxyl groups. In our study we show for the first time that this system can be also used to overcome the hydrophobic surface character of PCL fiber mats. To investigate the influence of the diamine concentration and of the coating time on the contact angle (CA), we added different amounts of HMD and chose coating times between 1h and 24h. We could show that the addition of HMD leads to a faster decrease of the CA and even with the lowest amine concentration a complete wettability could be achieved within 2h. Furthermore, we were able to show by EDX that the incorporation of HMD leads to an increase in the N-content. In addition results from SEM and first cell tests will be presented.

References

[1] D. Yan et al., *J. Med. Biol. Eng.* 33 (2013) 171-178.
[2] K. Fukatsu, *J. Appl. Polym. Sci.* 45 (1992) 2037-2042.
[3] S. Ku et al., *Biomaterials* 31 (2010) 9431-9437.
[4] Yang et al., *J. Mater. Chem. B* 3 (2015) 72-81.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P105

Biodegradable Inorganic Fiber Fleeces for Biomedical Applications

*B. Christ[1], S. Dembski[2,3], J. Probst[2], W. Glaubitt[2], H. Walles[3,1]
[1]Translational Center Würzburg ´Regenerative therapies in oncology and musculoskelettal diseases` branch of the Fraunhofer IGB, , Würzburg, Deutschland
[2]Fraunhofer Institute for Silicate Research ISC, , Würzburg, Deutschland
[3]Department Tissue Engineering and Regenerative Medicine (TERM), University Hospital Würzburg, , Würzburg, Deutschland

Tailored scaffold materials for the regeneration of damaged tissue or the construction of 3D tissue models continue to be in the focus of current research. Our focus in the development of novel scaffold materials is on inorganic oxide microfibers, which are processed to biodegradable fiber fleeces.

Viscous solutions are prepared out of liquid precursors, e. g. tetraethyl orthosilicate or tetraethyl orthotitanate via sol gel chemistry routes, and are spun under pressure to continuous filaments in an in-house developed spinning plant. This highly flexible technology allows not only to process different oxidic materials, but also to optimize material properties to the requested application area. Here, the fiber diameter, the (elasto)mechanical properties, the degradation rates in physiological media, the mesh sizes of the fiber fleeces (porosity) and even the orientation of the fibers within a fleece are adjustable.

Meanwhile, a wide range of application areas has been shown for these fiber fleeces. For instance, a silica gel fiber fleece has been CE-approved for the therapy of chronic skin diseases (diabetic ulcers and second-degree burns). After application of this biodegradable scaffold to chronic wounds, healthy cells grow into the fibrous matrix and, in parallel, the fibers are resorbed within about six weeks. Furthermore, this fiber fleece is used for the development and evaluation of economical and innovative therapy procedures for chronic skin diseases, which enable the patient to check on his/her own whether open wounds are healing. To test the effectiveness of medication, e. g. by integrating immuno-therapeutics into the fibers, artificial wound models are used at an early stage of development. This practice can significantly help to select effective medications for clinical tests. Moreover, by adding smart diagnostics in the form of targeted nanoparticles, specific biomarkers in the wound exudate can be imaged to visualize the proceeding of the healing process – in the future maybe via a specially equipped smartphone.

In summary, the authors present a technology for the production of sol gel based fibrous systems, whose property profiles can easily be adjusted to be used in *in-vivo*- and *in-vitro*-tissue engineering or even as drug carrier systems.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P106

Effect of simulated physiological solutions on ion release and apatite formation of bioactive glasses

*G. Kirste[1], J. Brandt-Slowik[1], D. S. Brauer[1]
[1]Friedrich-Schiller-Universität, Otto-Schott-Institut für Materialforschung , Jena, Deutschland

Tris buffer solution (Tris) and simulated body fluid (SBF) are standard solutions for testing the capability of bioactive glasses to precipitate apatite surface layers *in vitro.* Tris is commonly used for examining the early stage ion release at a physiological pH without interference of ions from the immersion medium except for chloride. The SBF"s composition on the other side mimics the much more complex inorganic part of blood plasm, including calcium, phosphate and carbonate ions among others. The apatite, which mineralizes on the surface of bioactive glasses, can show diverse substitutions. Chloride ions can be incorporated into the apatite lattice, forming chloro-apatites, and calcium ions can be replaced e.g. by strontium ions, which are efficient in the prevention and treatment of osteoporosis. The aim of this study was therefore to investigate the influence of different ions from Tris and SBF solutions on the apatite precipitation of bioactive glasses. For the purpose of examining the impact of chloride ions a conventional Tris, where hydrochloric acid is used to adjust the pH, was prepared as well as a chloride ion-free solution using acetic acid for pH adjustment. Besides both Tris buffer solutions three types of SBF where 0, 50 or 100% of Ca were replaced by Sr were used as immersion fluids for powder of Bioglass 45S5 for up to 7 days. X-ray diffraction, Fourier transform infrared spectroscopy and ^{31}P MAS NMR revealed significantly retarded apatite formation in the presence of strontium ions whereas chloride ions only slightly affected the rate of precipitation. In the absence of chloride ions calcite was precipitated in addition to apatite. ICP-OES analysis revealed significant differences in phosphate release for the different solutions, indicating that apatite formation is primarily limited by the phosphate supply owing to a change in glass dissolution in the presence of strontium ions or modified apatite solubility in the presence of chloride ions.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P107

Adipogenesis on geometrically defined 3D fiber scaffolds functionalized with decellularized adipose tissue (DAT)

*C. Blum[1], T. Schilling[2], K. Schlegelmilch[2], A. Ewald[2], T. Blunk[1], L. E. Flynn[3], J. Groll[2]
[1]University of Würzburg, Department of Trauma, Hand, Plastic and Reconstructive Surgery , Würzburg, Deutschland
[2]University of Würzburg, Department for functional materials in medicine and dentistry , Würzburg, Deutschland
[3]Queen's University, Department of Biomedical and Molecular Sciences , Kingston, Kanada

It´s known, that scaffold geometry triggers cell adhesion, differentiation and tissue ingrowth. *Melt Electrospinning Writing* (MEW) combines electrospinning of a polymer melt with computer-assisted moving of a collector to produce scaffolds of controlled geometry with precisely defined fiber thickness and mesh sizes.[1] The complex microenvironment of the extracellular matrix (ECM) is crucial to control stem cell differentiation.[2] While individual ECM components like collagen or laminin are commercially available, the native ECM provides many complex cues hard to reproduce artificially. Combining the 3D construct design with the adipose-inductive DAT promises to enhance adipogenesis. This project characterizes human bone marrow stromal cells (hBMSCs) response to DAT-functionalized 3D fiber scaffolds with or without exogenous adipogenic differentiation factors.

Materials and Methods

Poly(ε-caprolactone) (PCL) scaffolds were fabricated with MEW. DAT was adsorbed onto the scaffolds before seeding with primary hBMSCs. Adipogenesis was induced by differentiation factors in the culture medium, whereby different medium compositions were tested for up to 21 d. Induced and spontaneous adipogenesis on the scaffolds were analyzed by quantitative RT-PCR and Western Blot. Intracellular lipid droplets were visualized via Oil red O staining and quantified via trigylceride assay.

Results and Discussion

Adipogenic differentiation of hBMSCs on DAT-functionalized scaffolds as well as non-functionalized ones differed depending on the culture medium composition as detected by adipogenic mRNA and protein amounts. Moreover, fiber functionalization enhanced adipogenic outcome in corresponding media conditions. Oil red O staining and the triglyceride quantification of accumulated intracellular lipid droplets confirmed these findings as a function of the appropriate factor cocktail and fiber functionality.

Conclusion

The beneficial impact of fiber functionality on the adipogenesis of hBMSCs was proven. Defined scaffolds produced by MEW combined with fiber surface modification are a promising tool to further investigate the influence on hBMSC differentiation along the adipogenic lineage.

We thank the European Research Council (ERC) (consolidator grant Design2Heal, contract # 617989) and the German Research Foundation (DFG) (INST 105022/58-1 FUGG) for financial support.

References

[1] Brown T. D. et al., *Adv. Mater.* **2011**, *23*:5651.
[2] Badylak S. F. et al., *Acta Biomater.* **2009**, *5*:1.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P108

Permeability characterisation of marine-derived and synthetic bioactive glass based tissue engineered scaffolds

J. Hum[1], C. Daish[2,3], R. Blanchard[3], *A. R. Boccaccini[1], P. Pivonka[4]
[1]University of Erlangen-Nuremberg, Department of Materials Science and Engineering , Erlangen, Deutschland
[2]RMIT University, School of Engineering , Melbourne, Australien
[3]University of Melbourne, St Vincent's Department of Surgery , Melbourne, Australien
[4]Queensland University of Technology, Chemistry, Physics and Mechanical Engineering , Brisbane, Australien

Identifying suitable 3D scaffolds for repair and regeneration of bone in critical size defects is challenging due to different, often contradictory, requirements imposed on these materials. Ceramic materials have gained increasing use in bone tissue engineering due to their biocompatibility and similar chemical composition as bone. Particular bioactive glass (BG) patented under Bioglass 45S5, is still one of the most important biomaterials for bone defect repair. It was recently suggested that use of marine sponges as a sacrificial template for production of porous scaffolds may lead to better outcomes in terms of bone regeneration compared to using polyurethane (PU) foam templates. Recently, Boccaccini and co-workers have utilised marine sponges for generation of BG based scaffolds and characterised them in terms of compressive strength and effective diffusivity [1]. However, no assessment of scaffold permeability was performed.

This work investigates the permeability of new BG based tissue engineered 3D scaffolds. Permeability is directly linked to microstructural flow characteristics of scaffolds and may serve as a surrogate measure for blood vessel ingrowth. Scaffolds exhibiting low permeability may not be suitable for bridging of large bone defects due to hindrance of blood vessel ingrowth. The present work compares permeabilities of a novel type of BG scaffold derived from natural marine sponges with those from conventional scaffolds based on polyurethane (PU) foam templates. These scaffolds are generated using the foam replication method. It was previously shown [1] that one of the advantages of natural marine sponges is that they have a higher compressive strength (2-4 MPa) due to a decrease in porosity (68-76%) compared to the highly porous PU scaffolds with porosities around 90% and a lower compressive strength (~0.05 MPa).

Performing micro computed tomography (microCT) analyses of the generated 3D scaffolds at a resolution of 5 microns provides essential information on pore microstructure. The microstructures are imported into a finite volume software package for calculation of the pore scale fluid flow in different scaffold directions. Volume averaging of the velocity fields and pressure gradients allows to calculate the anisotropic permeability matrix.

References

[1] Boccardi et al. Oxygen diffusion in marine-derived tissue engineering scaffolds, J. Mater. Sci. Mater. Med. 26 (2015).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P109

Functional poly(2-alkyl-2-oxazoline)s for hydrogel formation and chemoselective coupling of biomolecules

*J. Blöhbaum[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, Lehrstuhl für Funktionswerkstoffe der Medizin und Zahnheilkunde , Würzburg, Deutschland

Poly(2-alkyl-2-oxazoline)s (POx) are a promising candidate for biomedical applications due to their good cyto- and hemocompatibility [1]. They are also very versatile due to their side chain variability [2]. Various monomers have been synthesized to introduce functional groups such as vinyl [3] or thiol functionalities [4].

The named groups can be used to perform thiol-ene chemistry for further functionalization or to produce hydrogels [5].

We are also interested in polymer-peptide conjugates synthesized by mild and chemoselective reactions. For this purpose, we described the introduction of cysteines to POx side chains via thiol-ene chemistry which could then be coupled to peptides via native chemical ligation [6].

A similar approach was used in this work to introduce thiols to POx side chains via post-polymerization functionalization. The amount of thiols was quantified via 1H NMR, Raman spectroscopy and Ellman assay. We also demonstrate hydrogel formation via thiol-ene chemistry and its potential application for 3D printing.

The thiol groups can further be used to introduce vinyl dimethylazlactone (VDM). VDM forms stable amide bonds with cysteine containing peptides in a mechanism similar to native chemical ligation and is stable under cell culture conditions [7]. This strategy could be an interesting alternative to the usually instable thioesters. Polymer-peptide conjugates were synthesized under mild conditions using VDM functionalized POX, which was confirmed by 1H NMR and HPLC measurements.

References

[1] Bauer, M.; Lautenschlaeger, C.; Kempe, K.; Tauhardt, L.; Schubert, U. S.; Fischer, D. *Macromol. Biosci.* 2012, *12*, 986.
[2] Hoogenboom, R. *Angew. Chem. Int. Ed.* 2009, 48, 7978 – 7994
[3] Gress, A.; Völkel, A.; Schlaad, H. *Macromolecules* 2007, 40, 7928
[4] Cesana, S.; Kurek, A.; Baur, M. A.; Auernheimer, J.; Nuyken, O. *Macromol. Rapid Commun.* 2007, 28, 608–615
[5] Stichler, S.; Jungst, T.; Schamel, M.; Zilkowski, I.; Kuhlmann, M.; Böck, T.; Blunk, T.; Teßmar, J.; Groll, J. *Annals of Biomedical Engineering* 2016, 45, 273-285
[6] Schmitz, M.; Kuhlmann, M.; Reimann, O.; Hackenberger, C. P. R.; Groll, J. *Biomacromolecules* 2015, 16, 1088−1094
[7] Schmitt, S. K.; Trebatoski, D. J; Krutty, J. D.; Xie, A. W.; Rollins, B.; Murphy, W. L.; Gopalan, P. *Biomacromolecules* 2016, 17, 1040−1047

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P110

Electrical fields in combination with matrix composition and stiffness control multipotent differentiation of human mesenchymal stem cells (MSCs) in an interdependent manner

R. Heß[1], A. Jäschke[1], H. Neubert[2], *D. Scharnweber[1]
[1]TU Dresden, Max-Bergmann-Zentrum , Dresden, Deutschland
[2]IKTS Dresden, , Dresden, Deutschland

Introduction

Cellular development is strongly dependent on numerous biochemical and physical signals provided by the cellular microenvironment. Traditionally, *in vitro* experimental studies concentrate on the influence of one signaling type and do not faithfully recapitulate the complex and dynamic signaling that predominates in vivo. The aim of this study was to determine how pure electrical AC fields – which have been already shown to stimulate osteogenic differentiation of human MSCs alone [1] – influence cellular commitment in combination with defined artificial extracellular matrices (ECM) of various stiffness.

Materials and Methods

For this, polyacrylamide (PAM) hybrid sandwich gels with stiffness ranging from 0.2 - 42 kPa were coated with collagen (coll) based matrices. Besides pure coll, matrices were altered by incorporation of hyualuronan (HA) and –derivatives varying in their degree of sulfation (sHA1; on average one sulfate group per disaccharide unit and sHA3; 3 sulfate groups per disaccharide unit). For electrical stimulation a new apparatus has been used applying AC electrical fields without interfering magnetic fields or biochemical reactions [1].

Results and Discussion

Electrical fields, biochemical composition and matrix stiffness alone affect cellular commitment. Particularly, sulfated HA-derivatives and increasing stiffness promote osteogenic differentiation while decreasing stiffness results in inhibition. Combination of selected matrix compositions / stiffness with electric fields further enhances osteogenic differentiation depending on the interplay between substrate stiffness and ECM composition. These date emphasize the complex and dynamic regulation of cellular commitment. An improved understanding of the interaction of these signals and an adequate control of stem cell differentiation into tissue-specific lineages will help to gather new insights into the osteogenic differentiation process and thus, improve current stem-cell based tissue engineering strategies.

Acknowledgements

We thank the German Research Foundation (Grant no: TRR67 TP A2/A3/Z3) for providing financial support to this project.

References

[1] Hess et al. (2012) *Cell Biochem Biophys* 64(3):223-32

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P112

Multifunctional nanostructured materials for hematopoietic stem cell culture

*A. Ludwig-Husemann[1], D. Kratzer[1], S. Kraus[1], C. Barner-Kowollik[2,3], C. Lee-Thedieck[1]
[1]Karlsruher Institut für Technologie, Institut für funktionelle Grenzflächen , Eggenstein-Leopoldshafen, Deutschland
[2]Queensland University of Technology, School of Chemistry, Physics and Engineering, Soft Matter Materials Group , Brisbane, Australien
[3]Karlsruhe Institute of Technology, Institute of Technical Chemistry and Polymer Chemistry, Dept. Of Polymer Chemistry , Karlsruhe, Deutschland

In the adult human body, hematopoietic stem cells (HSC) self-renew and give rise to all differentiated blood cells. Their function relies on a special microenvironment, called the HSC niche, which is found in bone marrow. Within the niche HSCs and their direct progeny sense this environment via cell surface receptors which enable cell-cell-interaction and cell-matrix-interaction, and recognize soluble factors. Furthermore the biophysical structure of the niche is essential for stem cell maintenance. Despite human HSCs have been successfully applied to reconstitute the hematopoietic system in the treatment of certain hematological disorders, the transplantation is still highly dependent of suitable donors and the fact of limited amount. To overcome this limitation the creation of an artificial niche mimicking the physiological archetype is an elegant tool to set up optimal conditions for the *ex vivo* expansion of HSCs. Such biomimetic niches should not only account for the biological and chemical factors present in the niche, but also for physical parameters such as nanopatterning.

To achieve this goal, the aim of the presented project is to develop multifunctional, nanopatterned cell culture substrates that allow directed functionalization with multiple biomolecules with special control over their arrangement on the nanometer scale. For this purpose, we test two different strategies: (i) the application of block copolymer micellar nanolithography to nanopattern growth factor binding hydrogels and (ii) usage of the breath figure technique to generate surface bound, orthogonal functionalizable, honeycomb-patterned porous films.

In the following, the effects on HSC behavior driven by the nature of the presented ligands, the ligand arrangement on the nanometer scale as well as the physical properties of the polymeric cell culture substrates will be investigated. The results of these studies will help us to understand and influence the control of HSC proliferation and differentiation.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P113

Crystallization of modified hydroxyapatite on titanium implants

*O. A. Golovanova[1]
[1]Omsk State University him F. M. Dostoevsky, Department of Inorganic Chemistry , Omsk, Russische Föderation

Introduction

Currently, much attention is paid to improving the quality of life and lifespan. In medical practice, to save lives and restore important physiological functions of patients we have to resort to reconstruction of bone and tooth tissue through replacing the defect with an implant. Since most of the bioactive materials lose their characteristics when attaching them to the bioinert (titanium) substrate, it is important to provide materials which exhibit high stability and retain their bioactive properties. In biomimetic synthesis, identification of optimal conditions for producing powders of highly dispersed phase with high resorption and biocompatibility with the human body are relevant.

Materials and methods

Synthesis of the modified hydroxyapatite was carried from the model medium similar to the human synovial fluid. Na2SiO3 was chosen as the basic compound, the modifier of the silicate groups, for the synthesis from the intercellular fluid model solution. To deposit hydroxyapatite on the VT1-0 titanium surface, the titanium plates 15 mm*15 mm*1.2 mm in size were pretreated.

Results and Discussion

The study of the surface and morphological characteristics of the produced phosphate coatings modified by silicate ions identified more complete deposition of Si-HA on the titanium substrate surface for etched samples. This treatment technique provides the coating which is uniform, dense, highly dispersed, and the HA crystals grow in the form of dendrites. The crystals tend to grow in a more structured form. Increase in the time of titanium soaking in the model sample solution leads to non-uniform growth of columnar-shaped crystals, which indicates the start of Si-HA surface structuring. After synthesis of the Si-HA layer on the titanium substrates, the samples were subjected to PIB. As a result, the coating was found to be uniform and dense with the edges fused. Crystals in the form of dendrites were observed on the substrates, which in our opinion, will contribute to further growth and renewal of the implant surface in physiological conditions. While determining the coated area, it was found that the highest percentage of deposition on the titanium substrate proceeds within the first three days. This is attributed to the fact that Si-HA dissolution in a static condition occurs mainly within three days, after which the dissolution of Si-HA is not observed.

Conclusions

The results of the study allow us to draw the following conclusions: all the samples synthesized in the model solution of the extracellular fluid under varying concentration of silicate ions are single-phase and represent hydroxyapatite; it is shown that the produced coating on the titanium is formed in several stages: growth in the form of the titanium crystal lattice; formation of crystals in the form of dendrites; crystal growth upwards in the form of cylindrical columns to form islands. *The work was carried out with the partial financial support of the Russian Foundation for Basic Research (grant No. 15-29-04839).*

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P114

Alginate hydrogels as versatile biomaterial for engineering hiPSC-derived cardiac cell models

*M. M. Gepp[1], A. Schulz[1], B. Fischer[1], J. C. Neubauer[1], H. Zimmermann[1,2,3]
[1]Fraunhofer Institute for Biomedical Engineering (IBMT), , Sulzbach, Deutschland
[2]Saarland University, Molecular and Cellular Biotechnology/Nanotechnology , Saarbrücken, Deutschland
[3]Universidad Católica del Norte, Facultad de Ciencias del Mar , Coquimbo, Chile

Human pluripotent stem cells are one of the most promising cell types for revealing diseases or developing novel therapies in regenerative medicine. Since the establishment of protocols for reprogramming somatic human cells into a pluripotent state (human induced pluripotent stem cells) [1], this field has further gained importance. In particular, patient-specific (e. g. cardiac disease related) pluripotent stem cells can be generated from skin biopsies and differentiated in vitro to cardiac cells for further applications of precision medicine (drug testing, drug discovery, etc.) [2]. Typical workflows comprise the expansion, differentiation and generation of final model. Especially the differentiated cells and models (neural, cardiac, etc.) require specialized cellular environments to reflect the real in vivo situation. Soft bioactive surfaces are in consequence replacing traditional stiff polystyrene surfaces because these are more comparable to the natural environment of the cells. It is still completely unknown how the "stem cell culture surfaces of the future" will look like, and thus, methods for chemical and topographical modification are highly required. The polysaccharide alginate (especially ultra-pure, xeno-free with low protein and endotoxin content [3]) is one promising biomaterial for engineering the required environments. We studied a couple of chemical and physical modifications of alginate hydrogels and developed bioactive alginate surfaces for adherent cultivation of both human multipotent and pluripotent stem cells [4]. Based on these findings, we further refined the alginate surface introducing a topography with small grooves (according to [5]). We present our recent work dealing with the long-term culture of hiPSC-derived cardiomyocytes on bioactive grooved alginate hydrogels.

References

[1] K. Takahashi, K. Tanabe, M. Ohnuki, M. Narita, T. Ichisaka, K. Tomoda, S. Yamanaka, Cell **131**, 861 (2007)
[2] I.Y. Chen, E. Matsa, J.C. Wu, Nature reviews. Cardiology **13**, 333 (2016)
[3] H. Zimmermann, D. Zimmermann, R. Reuss, P.J. Feilen et al., J. Mater. Sci.: Mater. Med. **16**, 491 (2005)
[4] M.M. Gepp, B. Fischer, A. Schulz, J. Dobringer, L. Gentile, J.A. Vásquez, J.C. Neubauer, H. Zimmermann, Journal of Applied Phycology, 1 (2017)
[5] T.G. Franzesi, B. Ni, Y. Ling, A. Khademhosseini, J. Am. Chem. Soc. **128**, 15064 (2006)

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P115

in vitro und *in vivo* biocompatibility analysis of a novel bone block consisting of collagen and a biphasic bone substitute as an alternative concept to allogeneic materials

*B. Woltmann[1], *R. E. Unger[2], J. Pissarek[1], S. Stojanovic[3], S. Najman[3], F. Witte[4], M. Barbeck[5,4]
[1]botiss biomaterials GmbH, , Zossen, Deutschland
[2]University Medical Center of the Johannes Gutenberg University, Mainz, Institute of Pathology , Mainz, Deutschland
[3]University of Nis, Faculty of Medicine , Nis, Serbien
[4]Charité - Universitätsmedizin Berlin, Julius Wolff Institute and Center for Musculoskeletal Surgery, Berlin-Brandenburg Center for Regenerative Therapies , Berlin, Deutschland
[5]botiss biomaterials GmbH, Research & Development , Zossen, Deutschland

Introduction

Composite materials made of *xenogeneic* collagen and alloplastic calcium phosphates might preclude the limited supply or additional risks known from autografts which are considered as gold standard. In the present study the cell- and tissue reactions to a new bone substitute material block made of synthetic biphasic hydroxyapatite/β-tricalcium phosphate (HA/β-TCP) granules (BCP) embedded in a naturally crosslinked porcine collagen matrix (maxresorb® flexbone) were analyzed *in vitro* and *in vivo*. A collagen containing cancellous allogeneic bone block (maxgraft®) and a synthetic biphasic bone block (maxresorb®) were used as controls. It was hypothesized that maxresorb® flexbone should exhibit regenerative properties comparable to allograft material.

Methods

An osteoblast cell line, MG-63, was used to evaluate the adherence and proliferation of the cells on the various biomaterials [1]. Cells were added and at specific time points after addition were examined morphologically and stained to determine distribution and expansion on the materials. Proliferation and cytotoxicity assays were used to evaluate and compare the growth rate of cells and cell compatibility of the various materials.

For the *in vivo* study the three materials were subcutaneously implanted in 45 rats for up to 60 days. Specialized established histological, (immuno-) histochemical and histomorphometrical methods were applied for analysis of the tissue response, i.e., involved cell types such as multinucleated giant cells (MNGCs) and implant bed vascularization [2].

Results

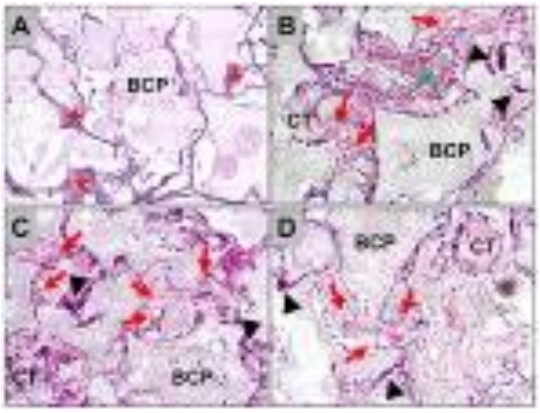
The *in vitro* results showed that the novel bone block led to a comparable adherence, cell morphology and proliferation of osteoblasts the allogeneic material and only slight differences were observed in the proliferation of cells on the various materials.

The *in vivo* results showed that the inflammatory tissue reactions, i.e., the numbers of MNGCs in case of the allografts and the novel bone block were comparable and significantly decreased compared to the numbers on the BCP group (Fig. 1). Moreover, the implant bed vascularization was superior to the BCP material alone and comparable to the allogeneic bone.

Conclusion

The results show that the novel bone block composed of BCP granules and native porcine collagen (maxresorb® flexbone) appears to be a favorable alternative to non-load bearing allogeneic bone grafts as both the regenerative potential and the integration behavior were similar.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P116

Scaffolds for off-the-shelf regenerative medicine

*B. Fischer[1], L. W. Weber[1], M. M. Gepp[1], A. Schulz[1], A. Meier[1], J. C. Neubauer[1], L. Gentile[1], H. Zimmermann[1,2]
[1]Fraunhofer IBMT, , Sulzbach, Deutschland
[2]Saarland University, Molecular and Cellular Biotechnology , Saarbrücken, Deutschland

In an aging society, calls for regenerative therapies increasingly rise. Besides the technical and biomedical hurdles, these calls can be addressed and realized only if economically acceptable. Especially for regenerative medicine, off-the-shelf solutions are a suitable strategy to create industrially robust cell-based therapeutics. Mesenchymal stem cells (MSCs) have shown great potential in regenerative medicine as they can be engrafted in a donor-independent way. Additionally, MSCs have been differentiated in a wide variety of therapeutically relevant cell types, *e.g.* bone, muscle and cartilage. Although MScs are capable of homing, degenerative pathologies may alter the niche, thus preventing a therapeutic effect of transplanted cells. Hence, a transplantable scaffold would not only help the cells to allocate properly, but offers them a physiological niche. Ultra-high viscosity (UHV) alginate (Alginatech, Germany) is a biopolymer derived from a unique blend of algae, which has shown great potential due to its biocompatibility and broad choice of biofunctionalizations. We devised a combinatorial cell-scaffold based system that relies on premade, application-dependent scaffolds, which simulate the niche to MSCs "on demand", thus inducing a specific and tunable lineage commitment (1). To evaluate the strength of commitment, our scaffolds were 3D printed with differing stiffness via a viscosity-independent method (2). In our experiments, MSCs were subjected to an open differentiation, where stiffness was the main differentiation cue and compared with MSCs exposed to an induced osteogenic differentiation. The commitment to a certain fate has been evaluated via gene expression profiling, principle component analysis and immunocytochemistry. Additionally, alkaline phosphatase activity has been analyzed to determine osteogenic induction. In conclusion, we showed that the stiffness of the substrate could recapitulate osteogenic differentiation. In the future, the combination of MSCs with off-the-shelf scaffolds can be used to expose cells to the physiologically relevant niche, without the need to specifically differentiate them in advance.

References

[1] A. J. Engler *et al.*, Matrix elasticity directs stem cell lineage specification. *Cell.* **126**, (2006).
[2] B. Fischer *et al.*, 3D printing of hydrogels in a temperature controlled environment with high spatial resolution. *Curr. Dir. Biomed. Eng.* **2**, (2016).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P117

Bone augmentation with biologically analogous mineral

*E. Hughes[1], R. Williams[1], S. C. Cox[1], M. Cooke[1], T. Hall[1], L. M. Grover[1]
[1]University of Birmingham, School of Chemical Engineering , Birmingham, Vereinigtes Königreich

Introduction- Intricate tubular channels found in bone (e.g. Haversian canals, Volkmans canals, and canaliculi), are integral for vasculature and cell communication processes that maintain bone viability. Methods used to synthesize commercial bone augmentation biomaterials produce highly crystalline materials that are absent of microstructures bone preserves. Here we describe calcium loaded hydrogel spheres that generate biologically analogous material within defects typical to those encountered in the clinical setting. In the presence of physiological fluids rich in phosphate, tubular structured mineral is deposited that augments the defect.

Methods- Calcium loaded spheres were made by adding 5 wt% agar powder to 1 M calcium nitrate solutions, before heating the mixture to 80-90 oC and feeding droplets of gel into a reservoir of liquid nitrogen. Deposition of tubular mineral was initiated by exposure to ammonium phosphate solutions at concentrations between 500 mM and 1 M, and was characterized by micro-XRF mapping, XRD and SEM techniques. For an *ex vivo* model, human bone tissue was collected from patients undergoing elective knee replacement surgery. The United Kingdom National Research Ethics Service (East of Scotland Research Ethics Service) provided ethical approval (11/ES/1044). The model was characterised by micro-XRF mapping and micro-CT techniques.

Results- Immersing calcium-loaded spheres in a phosphate reservoir promotes the release of calcium rich streams from the sphere surface, resulting in the precipitation of hierarchically structured low-crystallinity hydroxyapatite tubes, as determined by micro-XRF mapping, XRD and SEM. When brought into close proximity with one another, these spheres become fused in a matter of minutes through the entanglement and subsequent interstitial mineralisation of the mineral tubules. Micro-XRF mapping and micro-CT analysis of an augmented *ex vivo* human tissue defect model demonstrated the extensive deposition of low-crystallinity tubular mineral throughout the tissue defect.

Conclusions- This is possibly the first example of a bone augmentation material that is able to generate biologically analogous structures *in situ*, and therefore may serve as a better scaffold for bone formation over synthetic alternatives. Moreover, the formation of structured mineral aids in achieving rapid hardening of the augmenting calcium-loaded hydrogel within the defect space.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P118

Self-Assembly Mechanisms of Plasma Protein Hybrid Nanofibres of Albumin and Hemoglobin

*C. Helbing[1], T. Deckert-Gaudig[2], G. Wei[3], K. D. Jandt[1,4]
[1]Friedrich Schiller University Jena, Chair of Materials Science , Jena, Deutschland
[2]Institute for Photonic Technology, , Jena, Deutschland
[3]University of Bremen, Faculty of Production Engineering , Bremen, Deutschland
[4]Friedrich Schiller University Jena, Jena Center for Soft Matter , Jena, Deutschland

Protein nanofibers (PNFs) are fundamental buildings blocks of nature. Therefore, they are promising materials for applications in the field of biomedical engineering. PNFs consisting of plasma proteins, e.g. fibrinogen, albumin (HSA) and hemoglobin (HGB), are of special importance due to their biocompatibility. To expand the PNF application range and to achieve specific therapeutics goals, novel PNFs with multifaceted biological and physical characteristics are required. Therefore, the fabrication of novel hybrid protein nanofibers (hPNF) received increased attention because of their possibility of combining different protein functionalities and tailoring the resulting properties. Until now, there is no literature about the creation of PNFs consisting of two different plasma proteins. Therefore, we tested the hypothesis that the presence of HGB affects the self-assembly of HSA fibrils and will be incorporated to form the novel hPNF.

PNFs were self-assembled in a mixture of water and ethanol at elevated temperatures. The PNFs were characterized by atomic force microscopy (AFM), scanning transmission electron microscopy and tip-enhanced Raman spectroscopy (TERS) measurements. To determine the secondary structure of the proteins in solution during the fibre formation and their fibrillation formation kinetics circular dichroism spectroscopy (CD) was used.

We present novel self-assembled hybrid PNFs. A time-dependent structural polymorphism of hPNFs and composition-dependent fibrillation kinetics were demonstrated by AFM and CD measurements, respectively. The heterogeneous nature of the nanofibers was confirmed at the nanoscale resolution through TERS, due to the intrinsic molecular properties of HGB. Immunolabeling and force spectroscopy data corroborate the TERS results and indicate that HGB is being incorporated in the HSA nanofiber. We propose a dual-protein self-assembly mechanism model for the fibrillation of the hPNFs on the basis of the complementary results.

We demonstrate the possibility to create self-assembled PNFs in the presence of two different plasma proteins. Further, we confirmed the existence of a novel hPNF. These results lay the foundation for a novel biomaterial based on these hPNFs.

This work is part of the project: "Neue funktionelle Materialien basierend auf selbstassemblierten Protein-Nanofasern: Erzeugung und Verständnis von Nanofasern", AOBJ: 609403, which is funded by the Deutsche Forschungsgemeinschaft.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P119

Overcoming the hydrophilicity of bacterial nanocellulose for the transport of lipophilic drugs

*Y. Alkhatib[1], G. Blume[2], D. Kralisch[1,3], D. Fischer[1,3]
[1]Friedrich-Schiller-University Jena, Department of Pharmaceutical Technology , Jena, Deutschland
[2]Sopharcos, , Steinau, Deutschland
[3]Friedrich-Schiller-University Jena, Jena Center for Soft Matter (JCSM) , Jena, Deutschland

Question

The interest in new biomaterials for medical and pharmaceutical applications has dramatically increased in the last decades. The biopolymer bacterial nanocellulose (BNC), consisting of a unique three-dimensional network of nanosized fibers and over 90% water, has a great potential, due to its outstanding mechanical and biological properties. Although many attempts were made using BNC as a drug delivery system [1], loading with lipophilic substances is still a main challenge. In this study, entrapping the lipophilic model drug coenzyme Q10 (Q10) into the hydrophilic BNC network by innovative, dermal friendly and flexible colloidal carrier systems was investigated.

Methods

Strains of *Komagataeibacter xylinus* (DSM 14666) were employed to produce BNC fleeces in Hestrin-Schramm medium under static conditions in 24-well plates, which were harvested, alkaline purified [2] and optionally freeze-dried. As carrier systems to encapsulate Q10 Hydro-Tops (w/o/w nanoemulsion), Lipo-Tops (o/w emulsion) and liposomes were produced using a high pressure homogenizer. The stability of the prepared carrier systems regarding hydrodynamic diameter and zeta potential were measured up to 90 days at three different temperatures (4 °C, 22 °C, and 37 °C). The Q10 bearing carrier systems were loaded into BNC by a standard sorption method [2] and other post synthetic loading techniques such as injection and reswelling. Release was studied in purified water at 32 °C using the Franz cell diffusion system. Q10 was quantified by HPLC equipped with UV detector at 275 nm.

Results

Different carrier systems containing 0.5% Q10 were successfully prepared and revealed hydrodynamic diameters of about 65-130 nm, negative zeta potentials and an excellent stability over 90 days. BNC fleeces were efficiently loaded with these systems by the different post synthetic techniques. Q10 release could be controlled independency of the type of carrier systems, the BNC condition (native or freeze dried), and the type of loading technique.

Conclusions

The hydropolymer BNC could be successfully loaded with lipophilic substances, which opens a variety of applications. Moreover, variation of the parameters facilitates the adjustment of drug release for custom-designed application.

References

[1] S. Moritz, et al.; *Int. J. Pharm*., 471 (2014), pp. 45-55
[2] Y. Alkhatib, et al.; *Eur. J. Pharm. Biopharm*., 112 (2017), pp. 164-176

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P120

Release of TGF-β3 from Differently Modified Nanoporous Silica Nanoparticles for Cartilage Regeneration

*N. Schmidt[1], P. Behrens[1]
[1]Leibniz Universität Hannover, Institut für Anorganische Chemie , Hannover, Deutschland

Nanoporous silica nanoparticles (NPSNPs) have already offered their potential as delivery platforms in previous studies. The reasons are their outstanding properties including their high surface area, large pore volume (up to 50%), tunable particle and pore size and the amenability for surface modification. Moreover, NPSNPs have shown a good biocompatibility and are biodegradable.[1,2] Our approach is the application of such delivery systems for different growth factors on the surface of electrospun poly(ε-caprolactone) (PCL) fiber mats to build up a graded implant. This implant is applied in the case of an injured rotator cuff in the shoulder, offering a novel treatment for the clinical problem that after an injury the tendon-bone interface is often not regenerated.[3] The growth factors on the nanoparticle surface are supposed to stimulate the stem cells to differentiate and build up a regenerated tendon-bone connection after the degradation of the implant. As growth factors we used BMP-2 for the bone formation, TGF-β3 for the cartilage regeneration and Smad8 L-MH2 to stimulate the stem cells to form tendon cells. The presented work is focused on TGF-β3 and the cartilage regeneration. NPSNPs were prepared via a sol-gel process from alkaline aqueous solution. Cetyltrimethylammonium bromide (CTAB) was used as structure directing agent to build up the porous system.[4] The particle surface was modified with different trialkoxysilanes to equip the surface with various functional groups. We investigated the immobilization of TGF-β3 on the surface of the differently modified NPSNPs and its release. The immobilized and released amounts of TGF-β3 were determined by using an ELISA. Furthermore, the coating of the PCL fiber mat surface with NPSNPs was tested. The synthesized NPSNPs are approximately 40 nm in size and have a high specific surface area (500-1000 $m^2 g^{-1}$). The NPSNPs could be successfully used as delivery systems for immobilized TGF-β3. Moreover, the NPSNPs could be distributed nearly homogeneous among the whole surface of the PCL fiber mat.

This research project has been supported by DFG FOR 2180 "Gradierte Implantate für Sehnen-Knochen-Verbindungen".

References

[1] F. Tang et al., *Adv. Mater.*, **2012**, *24*, 1504-1534.
[2] Y. Wang et al., *Nanomedicine: NBM*, **2015**, *11*, 313-327.
[3] S. Font Tellado et al., *Adv. Drug Delivery Rev.*, **2015**, *94*, 126-140.
[4] Q. Huo et al., *Chem. Mater.*, **2009**, *21*, 3823-3829.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P121

Mesoporous Silica Nanoparticles for Drug Delivery in Head and Neck Cancer Therapy

*A. Watermann[1], S. Kurch[2], W. Tremel[2], J. Brieger[1]
[1]Universitätsmedizin Mainz, HNO-Klinik, Molekulare Tumorbiologie , Mainz, Deutschland
[2]Johannes Gutenberg Universität, Institut für Anorganische und Analytische Chemie , Mainz, Deutschland

Poor survival rates for head and neck squamous cell carcinoma (HNSCC) stagnate since decades due to limited therapeutic options, frequent recurrences and metastasis[1]. Therefore, it is of utmost importance to develop new therapeutic strategies. For example, targeted drug carriers such as dendritic mesoporous silica nanoparticles (MSNs) are highly promising[2]. MSN have a unique structure with an adjustable particle and pore size, are easily functionalized on the outer and inner surfaces, can be easily loaded with cargo and most importantly are highly biocompatible[3]. MSN are evaluated as drug delivery vehicles to head and neck cancer cells. An innovative gatekeeper system is linked to the MSNs" surface to encapsulate the drug and prevent its premature liberation.

MSNs showed uniform size and morphology. Cy-5 labeled MSNs were taken up by HNSCC tumor cells which was observed via flow cytometry and confocal microscopy. Also, metabolic activity was non-substantially reduced by particle concentration of up to 100 µg/mL. So, the MSN system is highly biocompatible and suited as a drug delivery vehicle for head and neck cancer therapy. Cancer cell specific uptake could be enhanced by coupling MSNs with an epidermal growth factor receptor antibody such as cetuximab.

References

[1] Robert Koch-Institut (Hrsg) und die Gesellschaft der epidemiologischen Krebsregister in Deutschland e.V. (Hrsg). Krebs in Deutschland 2011/2012; Berlin, 2015;
[2] Kienzle, A.; Kurch, S.; Schlöder, J.; Berges, C.; Ose, R.; Schupp, J.; Tuettenberg, A.; Weiss, H.; Schultze, J.; Winzen, S.; Schinnerer, M.; Koynov, K.; Mezger, M.; Haass, N. K.; Tremel, W.; Jonuleit, H. Dendritic Mesoporous Silica Nanoparticles for pH-Stimuli-Responsive Drug Delivery of TNF-Alpha. Adv. Healthc. Mater. 2017, 1700012, doi:10.1002/adhm.201700012.
[3] Vivero-Escoto, J. L.; Slowing II; Trewyn, B. G.; Lin, V. S. Mesoporous silica nanoparticles for intracellular controlled drug delivery. Small 2010, 6, 1952–1967, doi:10.1002/smll.200901789.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P122

Evaluation of ZnO nanoparticles for improvement of the irradiation response of tumor cells

*N. Wiesmann[1], J. Hill [1], M. Klünker [2], M. N. Tahir [2], W. Tremel [2], J. Brieger[1]
[1]Universitätsmedizin Mainz, HNO, Labor für Molekulare Tumorbiologie , Mainz, Deutschland
[2]Johannes Gutenberg-Universität, Institut für Anorganische und Analytische Chemie , Mainz, Deutschland

Radiation therapy plays a central role in cancer treatment, but unfortunately its success is limited by the development of resistances. Zinc oxide nanoparticles (ZnO-NP) were shown to exert selective cytotoxicity against tumor cells (1–3) most likely via the generation of reactive oxygen species (ROS) (4).

Our aim was to evaluate the applicability of ZnO-NP as radiosensitizer to improve the irradiation response of tumor cells.

We assessed tumor cell viability after treatment, the genotoxicity of ZnO-NP was analysed by γH2AX foci analysis and the performance of ZnO-NP as radiosensitizer was assessed by a colony formation assay.

We could demonstrate that ZnO-NP exert cytotoxicity to human tumor cells, which is conveyed by dissolved Zn2+ ions as well as by the particles themselves. Treatment with ZnO-NP resulted in double-strand breaks of DNA and the colony formation assay showed that treatment with ZnO-NP in combination with irradiation could enhance tumor cell death and reduce clonogenic survival.

We were able to show that ZnO-NP exert a genotoxic effect on human tumor cells. Combined treatment of tumor cells with ZnO-NP and irradiation with 2 or 4 Gray, according to typical, clinically applied irradiation dosages, resulted in reduction of tumor cell survival. All in all, the study shows that ZnO-NPs could probably be a promising anticancer agent, to improve the irradiation response of tumor cells.

References

[1] Chandrasekaran, M. et al. (2015) In Vitro Selective Anti-Proliferative Effect of Zinc Oxide Nanoparticles Against Co-Cultured C2C12 Myoblastoma Cancer and 3T3-L1 Normal Cells. Biological trace element research.
[2] Hanley, C. et al. (2008) Preferential killing of cancer cells and activated human T cells using ZnO nanoparticles. Nanotechnology, 19, 295103.
[3] Ostrovsky, S. et al. (2009) Selective cytotoxic effect of ZnO nanoparticles on glioma cells. Nano Res., 2, 882–890.
[4] Shen, C. et al. (2013) Relating cytotoxicity, zinc ions, and reactive oxygen in ZnO nanoparticle-exposed human immune cells. Toxicological sciences: an official journal of the Society of Toxicology, 136, 120–130.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P123

Use of amphiphilic polymers for controlled pharmacological release in drug eluting stents.

*S. Borrós[1], A. Ponce[1], V. Ramos-Pérez[1]
[1]Institut Químic de Sarrià, Universitat Ramon Llull, , Barcelona, Spanien

Introduction

During Percutaneous coronary interventions (PCI), the use of drug eluting stents (DES) have proven to reduce restenosis. Regardless of this evidence, good controlled pharmacological liberation is not always achieved although having a key role on endothelial healing. Therefore, the design of effective therapeutic strategies through polymer matrices is key factor to prevent restenosis after PCI. Some of the golden standards on DES to date are part of the lactic and glycolic families, which present low variation on drug release behaviour showing a limited tunability in their sustained release profile. In order to solve this issue, amphiphilic copolymers containing cholesterol, previously developed in our group [1], are proposed as an alternative. These show an excellent biocompatibility, a higher encapsulation ratio and can be easily formulated with other polymeric matrices to obtain a broad spectrum of releases allowing a better matching of the pharmacological requirements.

Materials & methods

Copolymers composed of a methacrylamide backbone and methacryloylated hydrophobic monomers derivatives of cholesterol are synthesized to use as suitable biocompatible coatings. Parameters such as drug load and drug/polymer ratio are enhanced to achieve different release profiles. Using a lab designed sprayer, different coatings are developed based on pHPMA and PLGA. These are checked by SEM and drug release is later characterized by HPLC.

Results

A 5% cholesterol pHPMA are synthesized and used successfully as a polymer matrix for new designed DES. Spraying parameters are optimized in order to obtain smooth morphologies and abluminal SEM images obtained from stents show good quality coatings. As seen in ***Figure 1***, releases in an accelerated medium range from 20% to 77% after 1h depending on the composition of the exterior layers. PHPMA exterior layers without pharmacological load show a slowdown in the overall release, being more intense when using PLGA. When the layers are formed by pHPMA containing drug, an initial burst is seen before 24h. This burst and barrier effects can be seen comparing groups S1-S4 or S5-S8

Conclusions

This combination of polymers enables a well defined and controlled drug release using an excellent biocompatible amphiphilic polymer. New stent designs are elaborated and characterized showing different release behaviours. Burst, barrier and sustained effects are obtained to enhance healing of the treated area.

Figure 1

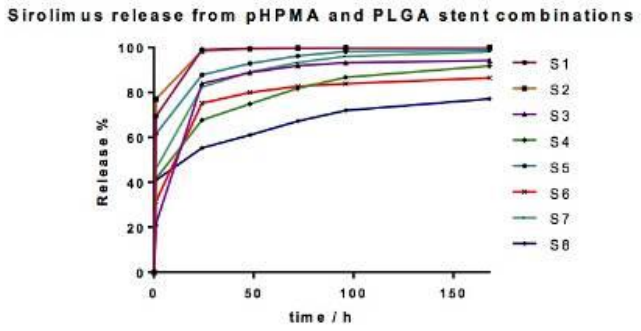


Figure 2

Stent	Nº of layers	Interior layers	Nº of layers	Exterior layers	mg/mm2
S1	10	pHPMA (siro)	-	pHPMA	
S2	10	pHPMA (siro)	10	-	0.53+-0.097
S3	10	pHPMA (siro)	10	PLGA	
S4	10	pHPMA (siro)	10	PLGA(siro)	0.68
S5	10	PLGA(siro)	-	-	
S6	10	PLGA(siro)	10	PLGA	0.45+-0.103
S7	10	PLGA(siro)	10	pHPMA	
S8	10	PLGA(siro)	10	pHPMA (siro)	0.92

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P124

Surface modification of drug delivery catheters using ND-YAG laser

*A. Thormann[1], M. Joseph[1], A. Heilmann[1]
[1]Fraunhofer-Institut für Mikrostruktur von Werkstoffen und Systemen IMWS, Technologien für Biofunktionale Oberflächen , Halle (Saale), Deutschland

Introduction

Drug-coated Balloon (DCB) catheters improve the performance of balloon catheters by delivering additional therapeutic effects to the treated vessels (Fig.1). The problem of poor integration drug on the surface and subsequent loss during insertion procedure itself limit its application in modern medical practice. This work introduces a laser surface micro-structuring technique to modify the catheter surface with drug carrying protective cavities. In application, open structures on inflated balloon surface efficiently fill the therapeutic agents and then deflated balloon surface carry and deliver drugs safely on desired application position.

Materials and methods

Catheter tubes which are made up of different medical polymers (TPU, PVC, PE and PEBAX) have outer diameters of 2.6 mm to 2.9 mm and wall thicknesses of 0.3 mm to 0.65 mm were used for micro-structuring. ATR-FTIR technique was used to figure out various photon absorption groups on the material surface and UV-Vis spectroscopy was used to detect corresponding absorption. A nanosecond Nd:YAG laser source of wavelength 355 nm and frequency of 15 KHz is selected for micro-structuring.

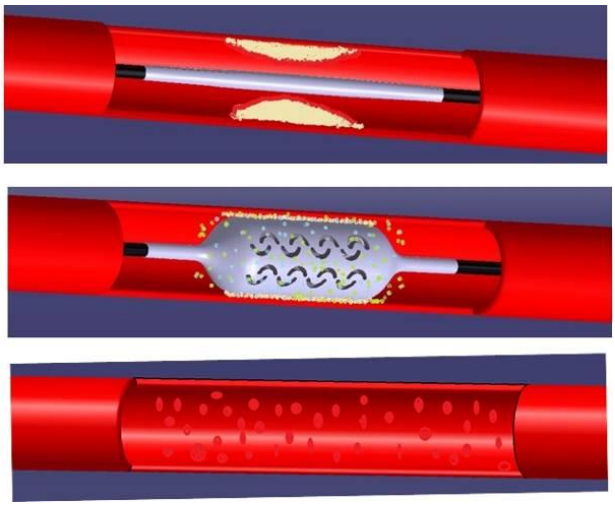
Results

The surface chemistry which was analyzed by ATR- FTIR spectroscopy shows the presence of good UV absorbers such as dienes (C=O), ketones (R-CO-R), amine (NH) groups on the all material surfaces except polyethylene. Moreover, the high peak intensity corresponds to low energy functional groups (CH) increases the hydrophobic behavior of materials. UV-Vis spectrum shows that almost constant absorption behavior for the spectrum of wavelength ranges from 300 nm to 1100 nm which increases the chance to select any of the three wavelengths (1064 nm, 532 nm and 355 nm) of Nd: YAG laser source for structuring. A 355 nm, 15 kHz laser source with various power irradiations (0.5 W to 1.49 W) and number of scans (2 to 20) have structured drug cavities of depth from 23 µm to 25 µm which was measured by light microscopy. The smooth, uniform structure which was observed by Scanning Electron Microscopy (SEM) helps to improve the efficiency of drug storage and delivery of drug coated catheters.

Conclusion

In conclusion, Nd: YAG laser source (355 nm, 15 kHz) can make good quality micro-structures on different catheter surface efficiently. The amount of material removal and quality of structures depends on material properties as well as various laser ablation conditions.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P125

Enzyme Degradable Polymersomes from chitosan-peptide-graft poly(ε-caprolactone) Copolymers

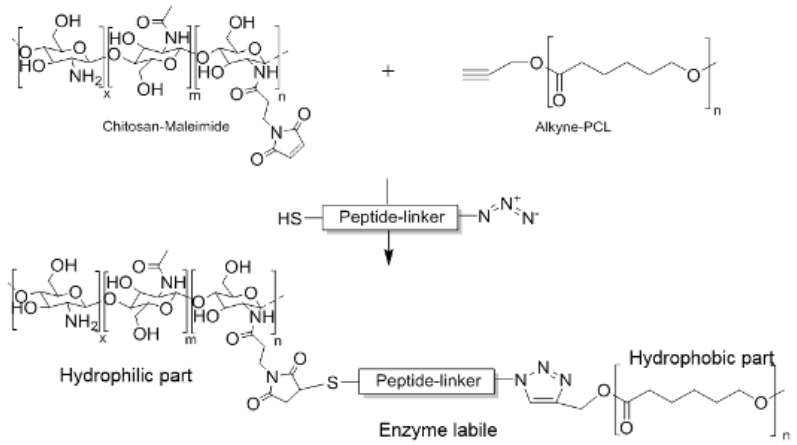
*Y. Bourgat[1], W. Tegge[2], B. Tiersch[3], H. Menzel[1]
[1]TU Braunschweig, Institut für Technische Chemie , Braunschweig, Deutschland
[2]HZI Braunschweig, , Braunschweig, Deutschland
[3]Universität Potsdam, , Potsdam, Deutschland

The long-term development and success of dental implants depends largely on the ability to keep them free of infections and with that to avoid any further surgical treatments. This research is designed to provide a pharmaceutical solution to prevent implant associated infections.[1] To face this challenge, a new amphiphilic graft-copolymer is introduced, which is based on two biocompatible materials such as chitosan (CS) and polycaprolactone (PCL). This graft-copolymer system is able to self-assemble into polymeric micelles via the solvent shift method. To make this graft-polymer responsive, the hydrophobic (PCL) and the hydrophilic (CS) parts are linked via a functional peptide.[2] The peptide is cleavable by an enzyme which is released as a result of the inflammation caused by the infection.[3] Antibiotics can be encapsulated in the micelles formed by the graft-polymer. A cleavage of the peptide by the enzyme will then lead to the separation of PCL and CS, and therefore in the disintegration of the micelles and a triggered release of the drug. The synthetic route involves the preparation of CS functionalized with maleimide groups (CS-mal)[4] and PCL with an alkyne end-group.[5] The peptide linker is equipped with a thiol and an azide end-group. Thiolen click-chemistry and an azide-alkyne Huisgen-cycloaddition are used to link the chitosan and the polycaprolactone chains, respectively, via the peptide.

References

[1] Pye, A.D., Lockhart, D.E.A., Dawson, M.P., Murray, C.A., Smith, A.J. J. Hosp. Infect. 2009 72(2):104-10
[2] Little C.B., Meeker C.T., Golub S.B., Lawlor K.E., Farmer P.J., Smith S.M., Fosang A.J. J. Clin. Invest. 2007; 117:1627–1636.
[3] Curtis CL, Rees SG, Little CB, et al. Arthritis Rheum 2002; 46(6):1544–53.
[4] X. H. He, L. Y. Liang, M. R. Xie, Y. Q. Zhang, S. L. Lin and D. Y. Yan, Macromol. Chem. Phys., 2007, 208, 1797–1802
[5] Chirachanchai S. et al. Macromol. Rapid Commun. 2016. 37, 1618−1622

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P126

Synthesis and Characterization of Polypeptoid Functionalized Silica Nanoparticles

*D. Nahm[1], B. Christ[1], S. Dembski[2], H. Walles[1], R. Luxenhofer[3]
[1]Würzburg branch of the Fraunhofer Institute Interfacial Engineering and Biotechnology, IGB, Translational Center Würzburg "Regenerative therapies in oncology and musculoskeletal diseases" , Würzburg, Deutschland
[2]Fraunhofer Institute for Silicate Research, ISC, Theranostic , Würzburg, Deutschland
[3]University of Würzburg, Functional Polymer Materials, Chair for Chemical Technology of Materials Synthesis , Würzburg, Deutschland

In recent years, nanoparticles for biological applications received a lot of attention in material science. Often, a polymer coating of these nanoparticles is used to ensure colloidal stability in biological media and to minimize or control the protein corona. Currently, polyethylene glycol (PEG) can be considered the gold standard used for this purpose.[1] However, PEG has recently been scrutinized and alternatives are being considered. With respect to biological aspects, a specific immune recognition of PEG and complement activation are being discussed.[2] Regarding the material science point of view, the major disadvantages result from the mechanism of the surface modification itself. Therefore, only a "grafting onto" approach is feasible usually leading to a so-called "mushroom regime".[3] This three-dimensional structure can result in insufficient surface coverage and in a poor accessibility of the polymer end groups.

Polypeptoids (poly(N-substituted glycine)s) are a class of biomimetic polymers which offer good solubility in water. Poly(N-methylglycin) exhibits excellent non-fouling properties and low cytotoxicity. This makes it an excellent choice for biomedical applications and an interesting alternative to PEG.[4] In addition, surface initiated polymerization or "grafting from" from non-colloidal surfaces with polypeptoids is already established and leads to high-density polymer brushes with excellent non-fouling.[5] Here, we report the synthesis of polypeptoid functionalized silica nanoparticles via surface initiated polymerization under various conditions. Primary amines, introduced by functionalization of the silica surface with one generation of a poly(amidoamine) dendron serving as initiating moieties. The prepared material was characterized by thermogravimetric analysis, dynamic light scattering, diffuse reflectance infrared fourier transform spectroscopy and zeta potential measurements.

References

[1]: Suk, J.; Xu, Q.; Kim, N.; Hanes, J.; Ensign, L.; *Adv. Drug Delivery Rev*., **2016**, *99*, 28.
[2]: Markovsky, E.; Baabur-Cohen, H.; Eldar-Boock, A.; Omer, L.; Tiram, G.; Ferber, S.; Ofek, P.; Polyak, D.; Scomparin, A.; Satchi-Fainaro, R.; *J. Controlled Release*, **2012**, *161*, 446.
[3]: Brittain, W.; Minko, S.; *J. Polym. Sci., Part A: Polym. Chem.*, **2007**, *45*, 3505.
[4]: Gangloff, N.; Ulbricht, J.; Lorson, T.; Schlaad, H; Luxenhofer, R.; *Chem. Rev.*, **2016**, *116*, 1753.
[5]: Schneider, M.; Fetsch, C.; Amin, I.; Jordan, R.; Luxenhofer, R.; *Langmuir*, **2013**, *29*, 6983.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P127

In vitro release of bone morphogenetic protein-2 from modified pcl fiber mats

*J. Sundermann[1], L. Burmeister[2,3], S. Sydow[4], D. de Cassan[4], H. Menzel[4], A. Hoffmann[2,3], H. Bunjes[1]
[1]Technische Universität Braunschweig, Institut für Pharmazeutische Technologie , Braunschweig, Deutschland
[2]Niedersächsisches Zentrum für Biomedizintechnik, Implantatforschung und Entwicklung (NIFE), , Hannover, Deutschland
[3]Medizinische Hochschule Hannover, Klinik für Orthopädie , Hannover, Deutschland
[4]Technische Universität Braunschweig, Institut für Technische Chemie , Braunschweig, Deutschland

Bone morphogenetic protein-2 (BMP-2) is a well-studied osteogenic growth factor that is, for example, utilized to induce spinal fusion in the authorized medical device INFUSE® Bone Graft. Due to its ability to induce new bone matrix, BMP-2 is mostly studied as a potential drug candidate for the repair of critical sized bone defects. A less common approach is to utilize BMP-2 to improve tendon-to-bone healing. This is based on the capacity of BMP-2 to encourage the differentiation of endogenous stem cells into osteoblasts which can support regeneration of the connective tissue between bone and tendon, the so called enthesis.[1] As BMP-2 has a short physiologic half-life after local administration, it is crucial to deliver the protein for a sufficient period of time.[2] Therefore, it is necessary to provide controlled release of the protein at the site of action while simultaneously ensuring its integrity and activity.

As an approach to the use of BMP-2 for tendon-to-bone healing it was incorporated in chitosan-tripolyphosphate-nanospheres, which were bound by electrostatic interactions to a multi-coated, electrospun PCL-fiber mat as a specifically designed implant-prototype. BMP-2 release studies confirmed a very efficient protein loading. The protein was released in a sustained manner for up to 28 days with initial burst release. Investigations by ELISA and BMP-Responsive Element (BRE)-Luciferase Assay[3] indicated that the released BMP-2 maintained its immunological integrity and biological activity over a period of at least 15 days.

References

[1] Lee, K-W. et al., J. Biomed. Mater. Res. Part B Appl. Biomater. 2016
[2] Ruhé, P. Q. et al., J. Mater. Sci.: Mater. Med. 2006, 17 (10), pp. 919–927.
[3] Korchynskyi, O. and ten Dijke, P., J. Biol. Chem. 2002, Vol. 277, 7, pp. 4883-4891.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P129

Polyglycidol Nanogels as Drug Delivery System as Strategy for Treatment of Type 2 Diabetes

*S. Hauck[1], T. Keller[1], T. Jüngst[1], S. Singh[2], A. Friedrich[3], K. Albrecht[1], H. Koepsell[3], J. Groll[1]
[1]University of Würzburg, Department for Functional Materials in Medicine and Dentistry and Bavarian Polymer Institute , Würzburg, Deutschland
[2]RWTH Aachen University, Institute of Technical and Macromolecular Chemistry , Aachen, Deutschland
[3]University of Würzburg, Department of Molecular Plant Physiology and Biophysics Julius-von-Sachs-Institute , Würzburg, Deutschland

Nanogels (NGs) are hydrophilic crosslinked polymeric particles that combine benefits of biocompatibility, high-water content and variable chemical and mechanical properties. These characteristics makes them attractive for delivery of large hydrophilic biomolecules like proteins and peptides. It has been shown, that a protein named RS1 induce downregulation of the glucose cotransporter SGLT1 by blocking exocytosis at the *Trans-Golgi-Network* (TGN). One RS1 domain, called RS1-Reg, regulates the release of SGLT1 containing vesicles from the TGN in a glucose dependent manner (Fig. 1). As a strategy for treatment of type 2 diabetes it can be possible to use RS1-Reg-derived peptides which upregulates SGLT1 at high glucose concentrations in small intestine. To provide the way of the peptides to the site of action in the cell and protect them against degradation we covalently coupled the cysteine-terminated peptides to a NG particle matrix through thiol oxidation during particle synthesis in the inverse miniemulsion. Due to the presence of the redox sensitive disulfide cross-links within the NGs, the particles are stable under acidic conditions in stomach but become mucoadhesive in the intestines. This behavior facilitates the contact of the peptide loaded NGs with the plasma membrane of the enterocytes. *In-vivo* tests in the murine model showed effective downregulation of SGLT1 after gavage of NGs loaded with a RS1-Reg derivate. This data implicates that the used NGs acts as an effective delivery system.

Figure 1

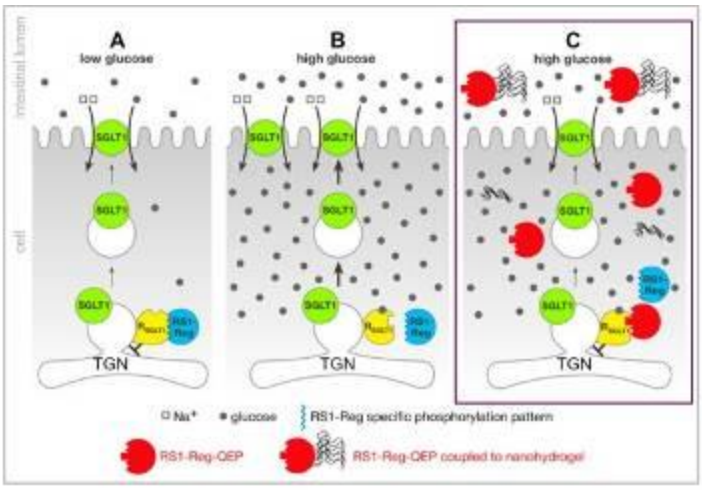


Figure 1. Model of the planed upregulation of SGLT1 in small intestine via with RS1-Reg variants coupled nanogels (Veyhl-Wichmann, M., et al. Molecular Pharmacology, 2016. 89(1): p. 118-132).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P130

Novel multifunctional thioether-polyglycidol coating for gold nanoparticles

*J. Lutz[1], S. Feineis[1], K. Albrecht[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, Abteilung für Funktionswerkstoffe der Medizin und Zahnheilkunde , Würzburg, Deutschland

Gold nanoparticles (AuNPs) are widely used due to the ease of their preparation in precise control over size and shape. After preparation, they are usually stabilized by charges or a tenside double layer, and further stabilization and biocompatibilization of the particles is usually achieved through ligand exchange reactions with thiofunctional molecules, often with thiol-terminated poly(ethylene glycol) (PEG-SH). Even though this is the established gold-standard, this procedure has disadvantages, since thiols are oxidation sensitive, and the presence of the highly nucleophilic thiols in molecules restricts the possibility to introduce other functional groups.

We have thus examined whether thioether may be used as alternative to thiols for stabilizing gold colloids. Here we present a systematic comparison of PEG-SH and PEG-thioether with multifunctional analogs, linear polyglycidol (PG) with multiple thiols (PG-SH) or ethylthioether (PG-SR) as coating system for AuNPs. We show that especially the multi-dental PG-SR displays outstanding colloidal stabilization, enabling lyophilization of such coated particles. Furthermore, the non-nucleophilic and non-oxidative character of thioether moieties provides the introduction of any functional mercaptan compound to allyl groups of the PG-SR backbone via thiol-ene click reaction.[1] In this manner a library of multifunctional PG-SR for AuNP coating was generated, featuring functionalities, such as charged moieties[2], biotin and diazirine moieties that can be used as generic tool for covalent immobilization of bioactive molecules.

References

[1] S. Stichler, T. Jungst, M. Schamel, I. Zilkowski, M. Kuhlmann, T. Böck, T. Blunk, J. Teßmar, J. Groll, Annals of Biomedical Engineering 2017, 45, 273.
[2] B. Baumann, T. Jungst, S. Stichler, S. Feineis, O. Wiltschka, M. Kuhlmann, M. Lindén, J. Groll, Angewandte Chemie International Edition 2017, 56, 4623.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P131

Synergism and Antagonism of Highly Hydrophobic Drugs Co-Incorporated into Polymeric Micelles

*M. Lübtow[1], L. Hahn[1], M. Salman Haider[1], R. Luxenhofer[1]
[1]Universität Würzburg, , Würzburg, Deutschland

High-throughput screenings can quickly assay the bioactivity of potential drugs, but when it comes to formulation, one major issue remains. According to estimates, more than 40% of all new chemical entities developed in the pharmaceutical industry are practically insoluble in water, underlining the urgent demand for excipients, which increase the water solubility without influencing the bioactivity of such hydrophobic drugs.[1] Motivated by the extraordinary high drug loadings of poly(2-oxazoline)s based micelles for paclitaxel (PTX) of more than 45 wt%[2], we investigated the specificity of a variety of pseudo-polypeptides based micelles for paclitaxel and curcumin (CUR) – both extremely water insoluble compounds (PTX: 0.4 mg/L[3]; CUR: 0.6 mg/L[4]). Depending on the polymer structure, we found pronounced drug-excipient specificities caused by minute changes in the carrier structure including structural isomers.

To avoid drug resistance after multiple injections of paclitaxel, the co-administration of curcumin seems to be beneficial.[5] In our case, the co–formulation of CUR and PTX with two structurally similar polymers showed very distinct drug-solubilization profiles. In the case of one polymer, we found synergistic drug loading, while it´s structural analogue suffered a clear antagonistic effect.

References

[1] E. M. Merisko-Liversidge, G. G. Liversidge, *Toxicol. Pathol.* **2008**, *36*, 43-48.
[2] a) R. Luxenhofer, A. Schulz, C. Roques, S. Li, T. K. Bronich, E. V. Batrakova, R. Jordan, A. V. Kabanov, *Biomaterials* **2010**, *31*, 4972-4979; b) A. Schulz, S. Jaksch, R. Schubel, E. Wegener, Z. Di, Y. Han, A. Meister, J. Kressler, A. V. Kabanov, R. Luxenhofer, C. M. Papadakis, R. Jordan, *ACS Nano* **2014**, *8*, 2686-2696; c) Y. Seo, A. Schulz, Y. Han, Z. He, H. Bludau, X. Wan, J. Tong, T. K. Bronich, M. Sokolsky, R. Luxenhofer, R. Jordan, A. V. Kabanov, *Polymer. Adv. Tech.* **2015**, *26*, 837-850; d) Z. He, X. Wan, A. Schulz, H. Bludau, M. A. Dobrovolskaia, S. T. Stern, S. A. Montgomery, H. Yuan, Z. Li, D. Alakhova, M. Sokolsky, D. B. Darr, C. M. Perou, R. Jordan, R. Luxenhofer, A. V. Kabanov, *Biomaterials* **2016**, *101*, 296-309.
[3] P. Ma, R. J. Mumper, *J. Nanomed. Nanotechnology* **2013**, *4*, 1000164.
[4] B. T. Kurien, A. Singh; H. Matsumoto; R. Hal Scotfield, *Assay Drug Dev. Technol.* **2007**, *5*, 567-576.
[5] S. Ganta, M. Amiji, *Mol. Pharm.s* **2009**, *6*, 928-939.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P133

Polyelectrolyte complex based on Carboxymethyl-kappa-carrageenan and Chitosan as prospective carriers for sustained antihypertensive drug delivery

*S. Lefnaoui[1,2], *M. M. Yahoum[1,2], *S. N. Gasmi[1], N. Moulai-Mostefa[2]
[1]1Faculty of Sciences University of Medea, Algeria, Department of Materials Science , Medea, Algerien
[2]2Materials and Environmental Laboratory, University of Medea , Medea, Algerien

1. Introduction. The matrices are carriers consisting of physiologically tolerated, inert excipients that do not disintegrate and form a network entrapping the active ingredient. Successful development and design of hydrophilic matrices is primarily determined by the selection of suitable excipients. Polyelectrolyte complexes (PECs) formation by the combination of opposing charge biopolymers proves to be a widely used method in the development of excipients for pharmaceutical use in the drug formulation. 2. Objective. Interpolyelectrolyte complexation between Carboxymethyl-kappa-carrageenan (CMKC), and Chitosan (CTS) was studied and characterized. PEC properties were determined by Fourier transform infrared spectroscopy (FTIR), X-ray diffraction and scanning electron microscopy (SEM). The PEC was used to develop matrix tablets aimed to sustain drug delivery. Valsartan matrix tablets were prepared by direct compression. Drug release in phosphate buffer solution medium (PBS, pH 1.2 and pH7.4) were reported. The dissolution data were fitted to different dissolution models. 3. Materials and methods. The PEC was synthesized and analyzed. Tablets containing Valsartan were prepared by direct compression. Various formulations based on PEC were prepared. The powder bed prior to compression was characterized for rheological properties according to standard procedures. The blended powder was then compressed into 400 mg tablets. The tablets were evaluated for the average weight, diameter, hardness value, friability percent, and drug content uniformity. The in vitro release studies of Valsartan from the prepared tablets were performed according to the USP30-NF25. The drug release data were fitted according to Korsmeyer–Peppas model. 4. Results. A PEC was formed by CTS and CMKC and confirmed by FTIR, DRX and SEM analyses. The rheological properties of the observed formulations indicated that the powder beds were suitable for compression and also good flowing. The drug release from matrix systems depends on the PEC concentration"s and approaches zero order kinetics. The results indicate that the release profiles could be controlled by modifying the proportion of PEC in the tablets. 5. Conclusion. A new PEC was prepared and characterized in order to evaluate its potential as sustained release matrix. PEC offered ideal excipients for novel tablet formulations with prolonged drug release.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P134

Formulation of wheat germ oil gelifieed emulsions stabilized by a microbial biopolymer

*M. M. Yahoum[1]
[1]university of Medea, Science of matter , Medea, Algerien

Introduction

Xanthan gum is a microbial biopolymer used as a thickener and viscosifying agent in food, pharmaceutical and cosmetic industries.

Objective

The aim of this work is to formulate a surfactifs free wheat germ oil (WGO) gelled emulsion stabilized with xanthan gum for use in the field of health as a dietary supplement, cosmetic cream or indirectly to increase the Effectiveness of certain active molecules (therapeutic adjuvant).

Materials and methods

Oil-in-water emulsions composed of 20% oil (WGO) and gelled with xanthan gum at different concentrations (0.5, 1, 1.5 and 2%) were formulated by direct method where both oily phase composed of WGO and gellified aqueous phase containing xanthan are first heated to a temperature of 60°C. Emulsification is then conducted by the addition of the oily phase portion wise to the aqueous phase under magnetic stirring and followed by a homogenization step. All formulations obtained were subjected to physicochemical controls, namely: Organoleptic proprieties accelerated and prolonged stability testing, microscopic controls, conductivity and viscosity measurements.

Results

The developed emulsions are all stable and exhibit a homogenous aspect and creamy appearance. The viscosity test showed that the products obtained exhibited a shear thinning behavior.

Conclusion

This study resulted in the development of a wheat germ oil-based emulgel stabilized by xanthan gum and totally exempted of synthetic surfactants. The products developed can therefore be considered as safe vehicles in pharmaceuticals or as cosmetic bases.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P135

Antibiotic-loaded bone allografts for prophylaxis and treatment of bone infections

*P. Donkiewicz[1], B. Buchbinder[1], S. Stojanovic[2], S. Najman[2], M. Barbeck[3,4], S. Wenisch[5], R. Schnettler[5]
[1]botiss biomaterials GmbH, , Zossen, Deutschland
[2]University of Nis, Faculty of Medicine , Nis, Serbien
[3]botiss biomaterials GmbH, Research & Development , Zossen, Deutschland
[4]Charité - Universitätsmedizin Berlin, Julius Wolff Institute and Center for Musculoskeletal Surgery, Berlin-Brandenburg Center for Regenerative Therapies , Berlin, Deutschland
[5]Universitätsklinikum Gießen, Klinik und Poliklinik für Unfallchirurgie , Gießen, Deutschland

Objective

Every surgical procedure is accompanied by an immanent risk of bacterial infections. This matter needs to be especially considered in transplantations of bone allografts for regeneration of lost tissue in dental and orthopedic applications, as the debridement is likely to result in worsening of the initial situation. One particular challenge is the population of bioimplant surfaces with biofilm forming bacteria such as *staphylococcus epidermidis* which are less prone to antibiotic treatment [1]. The loading of bone allografts with antibiotic agents prior to implantation demonstrates a promising approach to overcome this issue and prevent bacterial infections. In the present study both the pharmacokinetic properties of several antibiotics incorporated into allografts and their *in vivo* biocompatibility were analyzed.

Material and Methods

Freeze-dried bone allograft (FDBA) blocks (maxgraft®, botiss biomaterials GmbH, Berlin) were rehydrated in antibiotic solutions of either Clindamycin (lincosamide), Daptomycin (lipopeptide), Gentamycin (aminoglycoside), Rifampicin (ansamycin) or Vancomycin (glycopeptide) for 10 minutes in a 1:1 proportion prior to implantation. The concentration of remaining antibiotics was assessed every 24 hours after implantation with a total follow-up of 10 days. For analysis of the biocompatibility the antibiotic-loaded allografts were implanted into the proximal tibial bone and the initial tissue reactions were analyzed up to 3 days after implantation.

Results

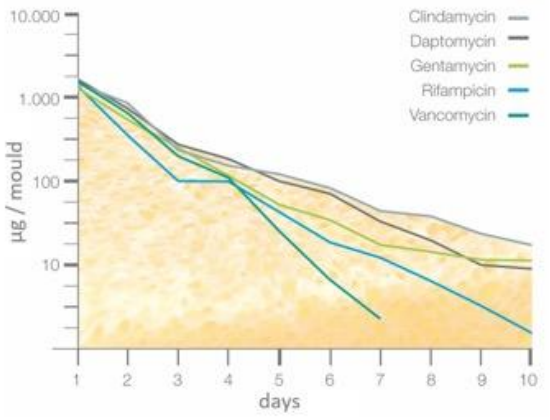
Antibiotics are easily incorporated into FDBA blocks and steadily released from the bone material for a period of over ten days within the analyzed time span (Fig. 1). Furthermore, the results of the *in vivo* study showed an excellent biocompatibility of all FDBA blocks combined with the different antibiotics.

Conclusion

The use of FDBA as carriers of antibiotic agents bares great potential in clinical application by eliminating systemic antibiotic-related side effects, minimizing risks of antibiotic resistance formation, providing advantages for eukaryotic cells for faster surface population and consequently minimizing the risk of surgical site infections and postoperative complications.

Fig. 1: Release kinetics of five antibiotic agents incorporated into allogenic bone blocks.

Figure 1



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P136

Tailoring thermal properties of biodegradable polyester based nanoparticles for drug delivery

*C. Helbing[1], D. Bandelli[2,3], C. Weber[2,3], U. S. Schubert[2,3], K. D. Jandt[1,3]
[1]Friedrich Schiller University Jena, Chair of Materials Science (CMS), Otto Schott Institute of Materials Research , Jena, Deutschland
[2]Friedrich Schiller University Jena, Laboratory of Organic and Macromolecular Chemistry (IOMC) , Jena, Deutschland
[3]Friedrich Schiller University Jena, Jena Center for Soft Matter , Jena, Deutschland

Biodegradable polymer nanoparticles (NP) are of great interest for drug delivery systems due to their outstanding biocompatibility and easy preparation. The major challenge for these systems is to tailor the degradation and drug release to the application they are aimed for. In the literature, it is reported that tuning the thermal properties of polyesters changes their degradation behaviour and the release kinetics of NPs. However, they did not include the variation of the hydrophilic / hydrophobic balance (HHB) and, thus, the encapsulation and release efficiency. Starting from this point, our aim is to create polyester based NPs with different thermal properties, whereby the HHB is kept constant. Here we present first results of polymer synthesis and NP formation.

Poly(ε-carpolactone) and two copolymers, one block- and one gradient copolymer, of Poly(δ-valerolactone) (PVL) and Poly(δ-decalactone) (PDL) were synthesized by ring opening polymerization. The thermal properties were measured by differential scanning calorimetry (DSC). NPs in aqueous suspension were prepared by nanoprecipitation from tetrahydrofuran. Dynamic light scattering was used to estimate the particle sizes. Individual single NPs were characterized by atomic force microscopy (AFM) in fluid and air as well as scanning electron microscopy (SEM).

The composition of the copolymers (20 mol% DL, 80 mol% VL) was carefully adjusted to match HHB of PCL. Kinetic studies performed during the statistical copolymerization revealed the gradient microstructure of PVL-*grad*-PDL and PVL-*block*-PDL copolymer. DSC measurements showed the presence of semicrystalline materials with a significantly lowered melting temperature of PVL-*grad*-PDL. Stable aqueous NPs suspensions of varying dimensions could be prepared by changing the concentration during the nanoprecipitation. NPs from all three polymers with a Dh of 170 nm remained stable for four weeks and were investigated in detail by AFM and SEM. All techniques consistently point towards an altered internal structure of the NPs with constant HHB.

These polymeric NPs will lay the foundation for a new type of polymeric drug delivery systems, which allows tailoring the degradation and release kinetics by changing their thermal properties.

This work is part of the Collaborative Research Center 1278: "Polytarget: Polymer-based nanoparticle libraries for targeted anti-inflammatory strategies" which is funded by the Deutsche Forschungsgemeinschaft (DFG).

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P137

Controlled Release of Alendronate for Local Delivery to Bone Tissue

*L. M. Grover[1]
[1]University of Birmingham, School of Chemical Engineering , Birmingham, Vereinigtes Königreich

Introduction

Alendronate (ALD) is used in clinical practice for the treatment of medical conditions characterized by extensive osteolysis. Recently local administration of ALD has been under investigation as a means to enhance bone fracture healing. The major issues encountered constitute the low encapsulation efficiency (EE%) and short release profile of ALD.

Objectives

This study aims to formulate a controlled-release delivery system for the local administration of alendronate to bone fractures via the formation of a polymer-mineral composite system.

Experimental methods

w/o/w double emulsion-solvent evaporation technique is used for the preparation of PLGA microspheres. Ca/P particles are prepared via a simple wet precipitation method following the Mobasherpour protocol (2007). Spectrophotometric evaluation of the samples is used to quantify ALD concentration. Microscopy is used to determine the size and morphology of the particles. Chemical analysis is performed via XRF and FTIR to identify the Ca/P phase of the mineral particles. Release studies were performed *in vitro* in distilled water. Preliminary toxicological evaluation is performed using the MC-3T3 cell line via the MTT assay. Student's t-test is used to distinguish statistically significant differences and the Thompson Tau analysis is used to determine outliers.

Results and discussion

Figure 1. (left) EE% of ALD loaded particles and (right) SEM image of HA-ALD loaded particles (scale bar 50μm). This chart summarises the impact of the particles" composition on the EE% of ALD. Both the increase in initial loading and the addition of Ca/P appear to improve the EE%. Comparison between micrographs a and b shows that the introduction of Ca/P particles in the formulation leads to less porous structures.

Figure 2. (left) Cell viability in the presence of PLGA particles, (right) SEM image of ALD loaded particles (scale bar 50μm). No toxic effect is caused by the introduction of particles in MC-3T3 cultures.

In vitro release experiments showed a decrease in burst release in the first 10days and also a prolonged release of ALD over a period of 70 days as a result of adding ALD as a Ca/P conjugate.

Conclusion

Efficient improvement of the EE% of ALD from the PLGA particles was accomplished by the introduction of Ca/P particles. Additionally the composite structures appear less porous which could support a more sustained and potentially controlled release of ALD which is observed in the release experiments. These structures proved to be bone cells" compatible based on no toxic effect observed.

Figure 1

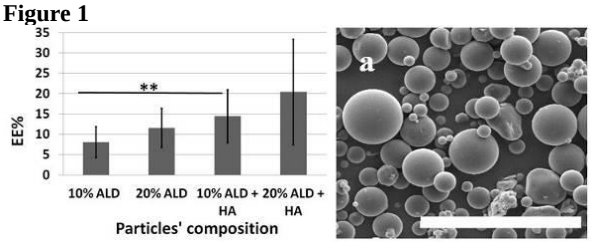
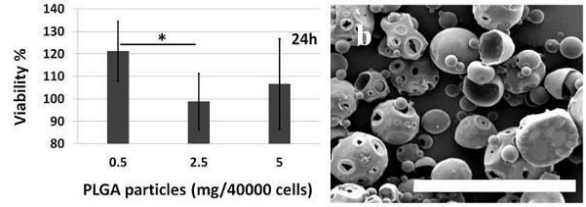


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P138

Collagen- silk fibroin hybrid nanofibrous membranes with fenugreek as bioactive wound dressings

*S. Selvaraj[1], D. Natarajan[1], M. S. Kiran[1], *N. Fathima[1]
[1]Central Leather Research Institute, Inorganic and Physical Chemistry , Chennai, Indien

Introduction

Antioxidants based wound dressings promote the rate of wound healing by reducing the free radicals, which hampers the normal healing process. Here, we propose collagen-silk fibroin hybrid nanofibers incorporated with fenugreek, an antioxidant, as a bioactive wound dressing material. These hybrid scaffolds with enhanced mechanical properties and biocompatibility can be an effective alternative to the conventional wound care products.

Methods

Nanofibers were prepared using co-electrospinning method. Morphology and mechanical properties of the nanofibers were evaluated using scanning electron microscopy and tensile tester. Antioxidant properties of the nanofibers were evaluated using 1, 1-diphenyl-2-picrylhydrazyl (DPPH) scavenging assay. Biocompatibility of the scaffolds was investigated using 3-(4,5-dimethylthiazol-2yl)-diphenyltetrazolium bromide (MTT) assay and the *in vitro* wound closure in the presence of release media was evaluated using mouse fibroblast 3T6 cell lines. Wound healing efficiency of fenugreek incorporated collagen-silk fibroin nanofibrous scaffolds was evaluated using full thickness excisional wounds in rat model.

Results

Tensile strength of the hybrid silk fibroin- collagen nanofibrous mats with fenugreek was higher than the collagen- fenugreek nanofibers. Fenugreek incorporated silk fibroin-collagen nanofibers showed 27 % DPPH scavenging activity. *In vitro* wound closure studies revealed that the wound closure is around 60% by 10 hours in the presence of release media whereas the untreated control cells show around 44% wound closure. The hybrid dressing showed higher wound healing rate in *in vivo* models than the untreated animals.

Discussion

In vivo wound healing studies revealed that the presence of phenolics in the fenugreek effectively reduces the free radicals generated during the inflammation phase of the wound healing. The presence of saponins in the fenugreek promotes the migration of fibroblasts thereby enhancing the wound closure in *in vitro* studies. Thus, these nanofibrous scaffolds could be ideal materials for wound dressings, where rich anti-oxidant environment is required.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P139

Biokompatibilität dünner, abbaubarer Beschichtungen zur Optimierung der Osteointegration bei gleichzeitiger Infektionsprophylaxe

*S. Burtscher[1], P. Krieg[2], M. Seidenstücker[1], A. Killinger[2], N. P. Südkamp[1], A. Bernstein[1]
[1]Universitätsklinikum Freiburg, Arbeitsgruppe des Labors für Muskuloskelettale Forschung , Freiburg, Deutschland
[2]Universität Stuttgart, Institut für keramische Bauteile , Stuttgart, Deutschland

Einleitung

Zu den häufigsten Ursachen für Implantatversagen in der Endoprothetik zählt die aseptische Prothesenlockerung, die auf das Fehlen der endgültigen Verankerung der Prothese in den umgebenden Knochen zurückzuführen ist. Auch die mikrobielle Besiedlung der Prothesenoberfläche und die damit verbundenen Infektionen stellen eine häufige Komplikation in der Endoprothetik dar. Ein Ansatz zur Reduktion dieser Komplikationen ist das Aufbringen von bioaktiven Substanzen auf Implantatoberflächen. Diese sollen sowohl antibakteriell als auch stimulierend auf das Osteoblastenwachstum wirken.

Fragestellung

12 dünne, abbaubare Beschichtungen, zusammengesetzt aus jeweils einer resorbierbaren, osteokonduktiven Keramik und einem bakterizid-wirksamen Metall sollten in Hinsicht auf ihre Biokompatibilität und bakteriostatische Wirksamkeit untersucht werden.

Methoden

Die Beschichtungen bestanden entweder aus Tricalciumphosphat (β-TCP), Hydroxylapatit (HA), GB14 oder Bioglas und waren jeweils mit Silber- , Kupferverbindungen oder Bismut (Bi) angereichert. Die Mischungen wurden mittels High Velocity Suspension Flame Spraying (HVSFS) in 20 µm Dicke auf Titanplatten aufgebracht. Zur Biokompatibilitätstestung wurden Bewuchsversuche mit der Zellkulturlinie MG63 durchgeführt. Histologisch wurde die Zellviabilität mittels Giemsa-Färbung und Live/Dead- Assay überprüft. Zur Quantifizierung der Zellvitalität in vitro kam das WST-1-Kit und zur Quantifizierung der Zytotoxizität, das LDH-Kit zum Einsatz. Eine Vorraussage über die Biokompatibilität in vivo wurde mittels Simulated Body Fluid erahnt. Durch Freisetzungsversuche wurde die Löslichkeit der Beschichtungen bestimmt und zur antimikrobiellen Testung wurde der Safe Airborne Antibacterial Assay mit S. aureus herangezogen.

Ergebnisse und Schlussfolgerungen

Die höchste Zellvitalität war bei β-TCP und Bioglas vorzufinden, bei GB14 waren die Ergebnisse durch die hohe Löslichkeit der Keramik nicht sicher einzustufen. HA zeigte besonders zu Beginn der Biokompatibilitätsversuche zytotoxische Wirkungen, die durch die Freisetzung toxischer Stoffe zu Beginn der Inkubationszeiten bedingt gewesen sein könnten. Hinsichtlich der antimikrobiellen Wirkung, zeigten Bi-Schichten im Unterschied zu Ag und-Cu Schichten keine Wirkung. Durch ihre hohe Löslichkeit und die hohe Freisetzung an Ionen, potenzierten die Bioglas und GB14-Keramiken die vorhandene antimikrobielle Wirkung des Cu und Ag.

Figure 1

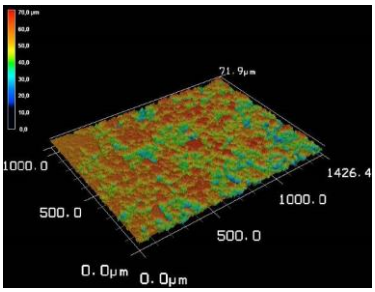
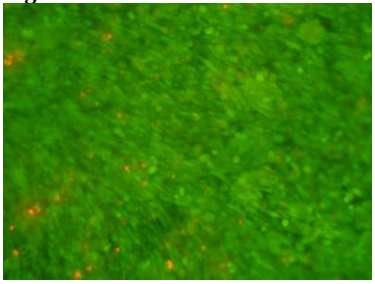


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P140

Platelet lysate – a serum alternative for human macrophage *in vitro* cultivation in mono- or co-culture with hBMSCs

*T. Tylek[1], T. Schilling[1], K. Schlegelmilch[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, ZMK/FMZ , Würzburg, Deutschland

Macrophages are cells of the innate immune system. They participate in phagocytosis of pathogens, the initiation of inflammatory reactions, as well as tissue repair and can be classified into inflammatory M1 and anti-inflammatory M2 macrophages. Together with multipotent human bone marrow stromal cells (hBMSCs), macrophages trigger tissue renewal and anti-inflammatory reactions [1]. Platelets are also part of the tissue regeneration processes. Once activated and degranulated they release different growth factors, which help to hence tissue repair. Because of this human platelet lysate (hPL) can be used as an alternative for serum component for cell culture [2]. Especially in the field of tissue regeneration and the corresponding use of biomaterials, hPL is becoming more and more interesting.

Here, we analyzed the performance of hPL as serum supplement for the *in vitro* co-culture of macrophages and hMSCs, which are commonly cultivated with different standard sera, i.e. human serum (hS) for macrophages and fetal calf serum (FCS) for hBMSCs. Via microscopy for the phenotype of both cell types, gene expression studies and the analysis of the phagocytic activity of macrophages after co-cultivation, we examined the differences in the use of hS, FCS, and hPL. We showed that in the co-culture, hPL leads to a comparable phenotype as in the standard culture sera. In addition, differences in gene expression and phagocytic activity were observed. Moreover, we demonstrated that hPL represents a successful alternative for macrophage mono-cultivation, since qPCR and flow cytometry proved spontaneous and induced differentiation into the different macrophage subtypes. As determined by the DNA amount cell adhesion in culture medium with hS and hPL was comparable and higher than in medium with FCS. The phagocytic activity of macrophages in hPL was approximately the same as in hS, but differed significantly from the results in medium with FCS.

With this data, we proved for the first time, that hPL is a successful alternative for macrophage mono- and co-cultivation with hMSCs, especially overcoming the negative effects of FCS on macrophages. Thereby, both cell types show comparable phenotype and functional characteristics to their respective standard sera in cell culture.

References

[1] Eggenhofer & Hoogduijn; Transplant Res; 2012
[2] Burnouf et. al; Biomaterials; 2016

We thank the European Research Council (ERC, consolidator grant Design2Heal, contract no 617989) for funding.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P141

The interaction of ultra-small gold nanoparticles with bacteria - an imaging study

*N. Bialas[1], T. Ruks[1], V. Sokolova[1], M. Epple[1]
[1]University of Duisburg-Essen, Inorganic Chemistry and Center for Nanointegration Duisburg-Essen (CeNIDE) , Essen, Deutschland

Introduction

The interaction of metallic nanoparticles with microorganisms is of great importance in biomedicine. Particle size is the essential parameter which determines the capability of metallic nanoparticles to be taken up by cells. Gold nanoparticles may serve as carriers for biomolecules. Fluorescence microscopy is a powerful tool to investigate the interaction of metallic nanoparticles with bacteria and demonstrate whether it has an extra- or intracellular nature.

Objectives

The aim was to study the particle-cell interaction and the possible uptake of ultra-small gold nanoparticles with bacteria. The nanoparticles are small (2 nm) compared to the bacteria (0.5-3 µm).

Materials and methods

Ultra-small gold nanoparticles were synthesized by reduction of gold ions (HAuCl4) with NaBH4 in the presence of the capping agent glutathione (for red fluorescence) and fluorescently labeled hexapeptides (for green fluorescence) via cysteine coupling. Afterwards they were purified by centrifugation and characterized by differential centrifugal sedimentation, 1H-NMR spectroscopy and transmission electron microscopy. *Escherichia coli* DH5α and *Staphylococcus xylosus* DSM 6179 strains were transformed with a plasmid vector pGEX6P1 encoding the green fluorescence protein (GFP). GFP expression in bacteria was induced by the addition of isopropyl β-D-1-thiogalactopyranoside to log phase cultures. Green-fluorescent strains were then incubated with red-fluorescent gold nanoparticles (autofluorescence), whereas green-fluorescent gold nanoparticles were incubated with *Escherichia coli* TOP10 strain which, due to the expression of DsRed2 protein in cells, demonstrated red fluorescence. The interaction of nanoparticles with bacteria was studied by fluorescence microscopy and confocal laser scanning microscopy.

Results

Gold nanoparticles had spherical morphology with an average size of 2 nm. Bacterial strains were successfully transformed with the GFP-encoding vector. The interaction of nanoparticles with bacteria was demonstrated in all strains by following their fluorescence.

Conclusions

E. coli DH5α and *S. xylosus* DSM 6179 strains underwent efficient transformation with the GFP-encoding vector. All bacteria demonstrated bright fluorescence. By fluorescence microscopy, the interaction of fluorescent nanoparticles with bacteria was shown.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P142

Melt electrospinning writing of tailored sinusoidal elastomer fiber scaffolds for tendon and ligament repair

*G. Hochleitner[1], F. Chen[2], C. Blum[1], B. Amsden[2], P. D. Dalton[1], J. Groll[1]
[1]Universitätsklinikum Würzburg, Lehrstuhl für Funktionswerkstoffe der Medizin und Zahnheilkunde , Würzburg, Deutschland
[2]Queen's University, Department of Chemical Engineering and Human Mobility Research Centre , Kingston, Kanada

Introduction

Tendon and ligament tissue is composed of crimped collagen fibers with a specific stress-strain behaviour.[1] To mimic this, photo-cross-linkable copolymers made of L-lactide (LLA), ε-caprolactone (CL) and acryloyl carbonate (AC) were synthesised and printed via melt electrospinning writing (MEW).[2] Resulting defined sinusoidal structures showed the characteristic "toe region" of load bearing soft tissue. The scaffolds were cytocompatible and exhibited an elastomeric performance, including a high elasticity, strength and creep-resistance.

Methods

p(LLA65-ε-CL25-AC10)[3] and p(ε-CL90-AC10) were synthesised and characterised (GPC/NMR/DSC). For cross-linking, polymers were mixed with 1 wt.% I651. For MEW, a customized device has been set up (Figure 1A).[4] By variation of the pressure and collector speed, different patterns were deposited (Figure 1B). Subsequent tensile testing with and without additional preloading as well as eluate assays with L929 fibroblasts and adhesion tests with hMSCs were conducted.

Results & Discussion

Straight p(ε-CL90-AC10) fibres (fØ ≈ 30 µm) revealed a maximum tensile strength σ=53±16 MPa, Young"s modulus E=314±157 MPa and ultimate strain ε=90±12 %. By reducing the collector speed, differently shaped sinusoids were printed with reduced stiffness, E=29±17 MPa. Mechanical testing revealed a high creep resistance and elasticity (Figure 2) with and without preloading (n=10^4, 1 Hz, 10 % strain). After 7 days, vital hMSCs covered the scaffold surfaces.

Conclusion

MEW of tailored sinusoidal elastomer fibers is a promising tool for printing biocompatible scaffolds. These are highly elastic, creep-resistant and strong, in contrast to commonly used thermoplastics for tissue engineering. The customized "toe region" enables to mimic the mechanical behaviour of tendons or ligaments.

Acknowledgement

The authors acknowledge the funding of Bone and Joint Health CREATE Program, Discovery Grant salary support and the European Research Council consolidator grant Design2Heal.

References

[1] C. K. Kuo, Sports Med Arthrosc Rehabil Ther Technol, 2 (2010), 20
[2] T. D. Brown, Adv. Mater., 23 (2011), 5651–5657
[3] F. Chen, Biomacromolecules, 17 (2016), 208−214
[4] G. Hochleitner, Polymer, 55 (2014), 5017-5023

Figure 1

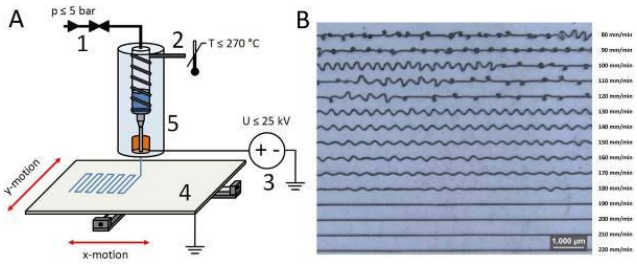
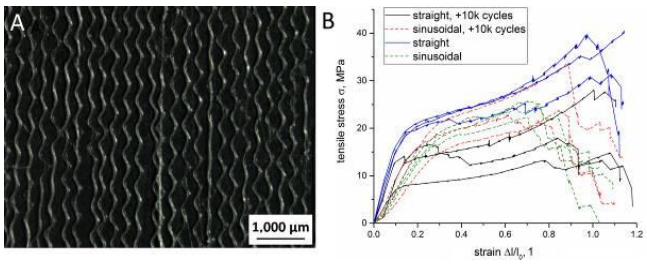


Figure 2



Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P143

Tissue engineered pre-vascularized buccal mucosa equivalents utilizing a primary triculture of epithelial cells, endothelial progenitor cells and fibroblasts

*M. Heller[1,2], H. K. Bauer[1,2], E. Schiegnitz[3,2], R. Schwab[1,4], A. Hasenburg[1], R. E. Unger[5,2], W. Brenner[1,2]
[1]University Medical Center of the Johannes Gutenberg University Mainz, Department of Gynecology , Mainz, Deutschland
[2]University Medical Center of the Johannes Gutenberg University Mainz, For BiomaTiCS – Biomaterials, Tissues and Cells in Science , Mainz, Deutschland
[3]University Medical Center of the Johannes Gutenberg University Mainz, Department of Maxillofacial Surgery , Mainz, Deutschland
[4], ,, Deutschland
[5]University Medical Center of the Johannes Gutenberg University Mainz, Department of Pathology , Mainz, Deutschland

The management or reconstruction of bigger defects after tumor surgeries is an issue of great interest, especially in areas in which cosmetically appealing results are desired. After tumor resections of the vulva, vagina or the mamma, there are extensive reconstructive surgeries necessary using implants or flaps. Besides the disadvantages after transplantation such as comorbidity of a second trauma, pain and scar formation, one of the biggest issues is a poor blood vessel supply, resulting in an increased morbidity and necrosis. One promising alternative to the commonly used wound coverage approaches in reconstructive surgery is the use of artificially generated pre-vascularized soft tissue equivalents. In our group we generated a pre-vascularized buccal mucosa equivalent in a tri-culture of primary buccal epithelial cells, fibroblasts and endothelial progenitor cells, using a native collagen membrane as a scaffold. A successful pre-vascularization and dense formation of capillary-like structures at superficial areas of the matrix was demonstrated. Additionally, we found a distinctly increased formation of capillary-like structures on the collagen membrane. In *in vivo* studies the functional connection of these capillary-like structures was demonstrated by the formation of anastomosis with the host vasculature after implantation in nude mice. Pre-vascularized buccal mucosa equivalents were generated successfully and may lead to accelerated and improved blood vessel supply of the transplanted tissue equivalents resulting in higher success rates after transplantation.

Abstracts – Annual Meeting of the German Society for Biomaterials 2017 – Würzburg, November 09 – 11 • DOI 10.1515/bmt-2017-6016
Biomed. Eng. - Biomed. Tech. 2017; 62(s2): p90-p228 • © by Walter de Gruyter • Berlin • Boston

P144

3D Structured Tissue Scaffolds for High Resolution Microscopy

*S. Mayr[1], B. Buchroithner[1,2], B. Buchegger[2], J. Jacak[1]
[1]University of Applied Sciences Upper Austria, School of Applied Health and Social Sciences , Linz, Österreich
[2]Johannes Kepler University, Department of Applied Physics , Linz, Österreich

Tissue engineering is a rapidly growing field. Often, cells within an artificial tissue need structural support or guidance for growth which can be provided by a polymeric scaffold. The fabrication of arbitrary, bio-compatible scaffolds can be accomplished by multi-photon lithography (MPL).

In MPL, a femtosecond-pulsed laser focused into a liquid, photosensitive resin solution initializes polymerization solely within the focal volume of the laser beam. Hence, sub-micrometer resolution can be achieved in three dimensions.

Using MPL, we are able to print 3D structured polymer scaffolds for cells and tissue with a resolution below the diffraction limit. The challenge herein is the development of a photoresist that is biocompatible, mechanically stable and can be structured at a high writing speed. Methacrylates - in comparison to acrylates - provide a good biocompatibility for cells. However, methacrylates have a lower cross-linking reactivity. To speed up the fabrication process, we employ radical thiolene polymerization (addition of a tetra-functional thiol-group named pentaerythritol tetrakis mercaptoacetate). Moreover, a low percentage of trifunctional acrylate is mixed into the photoresist.

So far, we achieved biocompatible, three-dimensional polymer scaffolds with a writing speed greater than 100 µm/second. For biocompatibility testing the scaffolds are seeded with cells and their viability is proven with an established viability assay. In order to promote cell adhesion, we developed strategies to functionalize the scaffolds with biomolecules like RGD-containing peptides or proteins via click-chemistry and chelation reactions, respectively.