Leveraging additive manufacturing for advancing the fabrication of next-generation in vitro 3D tumor models with personalized hierarchical features, anatomic size and defined morphology is highly advantageous for recapitulating the cellular and extracellular building blocks of the tumor microenvironment from the bottom-up [1]. The development of such biomimetic testing platforms with anatomic length scales unlocks new avenues to program cancer-stromal cells spatial distribution and introduce tumor-ECM components in a user-defined mode that will contribute for accelerating preclinical drug screening and validation. To engineer such organotypic human-sized tumor models, herein we take advantage of freeform suspension 3D bioprinting using continuous viscoelastic supporting baths comprising cost-effective food thickening polysaccharides [2] and tumor-ECM mimetic photo-crosslinkable bioinks laden with key components found in human tumors. This approach enabled the manufacture of shape-defined, cell-rich, 3D multi-layered tumor constructs with high resolution. Malignant and cancer cells remained viable, proliferated within 3D bioprinted ECM-mimetic constructs and secreted key growth factors (e.g. TGF-β, FGF-2). The biofabricated 3D tumor models were highly reproducible and suitable for screening anti-tumoral therapeutics. Overall, the developed approach is highly programable from the bottom-up and transversal, allowing the manufacture of a myriad of anatomically defined 3D tumor testing platforms for screening candidate anti-cancer therapeutics bioperformance in a preclinical in vitro setting.

Keywords: Freeform 3D Biopritning; Tumor Models; Preclinical Drug Screening

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Abstract 1606

INNOVATIVE MATERIALS FOR THE PREVENTION OF POST-SURGICAL ADHESION

Franka Ennen-Roth[1], Abdolrahman Omidinia Anarkoli[1], Andreas J. Krüger[1], Laura De Laporte[2,3,1]

[1]*DWI - Leibniz Institute for Interactive Materials, Aachen 52074, Germany*
[2]*Department of Advanced Materials for Biomedicine, Institute of Applied Medical Engineering, RWTH Aachen University, Aachen 52074, Germany*
[3]*Institute of Technical and Macromolecular Chemistry, RWTH Aachen University, Aachen 52074, Germany*

Corresponding author's email: ennen@dwi.rwth-aachen.de

The AparTex project focusses on the development of a novel absorbable barrier material to prevent tissue adhesion after surgical interventions.

Adhesion of organs and adjacent tissue after a surgery effects up to 95% of patients during the healing process. This minimizes life quality, can lead to loss of organ function, and displays a huge financial burden on the health care system.

Established anti-adhesion materials used as temporary barriers exhibit several drawbacks concerning flexibility, stability, functionality, or handling.

Customized electrospun materials made from functional hydrogel fibers aim to improve the post-operative wound healing and patient recovery.

While one layer of the AparTex textile would stick to the injured tissue, the other layer of the textile will keep the healthy tissue at a distance and act as a physical, absorbable barrier.

Here, we present first prototypes and their properties in terms of mechanical properties, degradation properties and biocompatibility.

Keywords: adhesion; electrospinning; fiber

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Abstract 1607

ECM MIMICKING FIBROUS AND VISCOELASTIC SUPRAMOLECULAR HYDROGELS FOR TISSUE ENGINEERING AND 3D PRINTING

Shahzad Hafeez[1], Antonio Feliciano[1], Agustina Aldana[1], Nicholas M. Matsumoto[2], René P. M. Lafleur[2], Lorenzo Moroni[1], Clemens C.V. Blitterswijk[1], Bert E.W. Meijer[2], Matthew B. Baker[1]

[1]*Department of Complex Tissue Regeneration (CTR), MERLN Institute, Maastricht University, the Netherlands*
[2]*Institute of Complex Molecular Systems (ICMS), Eindhoven University of Technology, Eindhoven, the Netherlands*

Corresponding author's email: s.hafeez@maastrichtuniversity.nl

Traditional hydrogel lacks fibrous structure, dynamicity and viscoelasticity found in native extracellular matrix (ECM). ECM is the non-cellular component present within all tissues and consists of fibrous structures like collagen, laminin and fibrin. Nature exploits self-assembly and constituents interact via non-covalent interactions to form superstructures found in ECM. Despite recent advances, creating synthetic analogues of ECM with fibrous structure and controlled viscoelasticity remain a challenge. We have explored the use of 1,3,5-benzenetricarboxamide (BTA) based hydrogelators as synthetic mimics of the fibrous ECM for tissue engineering. First, via modular mixing of BTA, we created supramolecular hydrogels with controlled superstructures and viscoelasticity. BTA hydrogels are highly cytocompatible and can direct cell-cell interactions based on their dynamics and mechanical properties. We then developed a desymmetrization method to upscale and shorten BTA synthesis route. With desymmetrization, molecular control has made possible the development of new fibrous architectures, which are injectable, extrudable and show good shape fidelity after 3D printing. Another challenge is the erosion of highly dynamic BTA hydrogels over time in cell culture. We address this by making BTA monomer with norbornene on it. This newly developed hydrogelator form micron-long fibers. These fibers are polymerized using thiolene chemistry, resulting in tougher and stable hydrogel. This hydrogelator showed high cell viability, very good extrusion and amenable to fabrication via digital light processing. These newly developed hydrogelators opens up numerous applications in tissue engineering, and remain one of the few examples of synthetic and tunable supramolecular hydrogels platforms that show applications in 3D printing.

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Abstract 1608

SUCCESSFUL ISOLATION AND CULTURE OF MULTIPOTENTIAL DISTAL AIRWAY STEM CELLS FROM COPD PATIENTS

Tina Patricia Dale[1], Michael Santer[1], Sana Iftikar[2], Mohammed Haris[2], Nicholas Robert Forsyth[1]

[1]*Keele University*
[2]*Royal Stoke University Hospital*

Corresponding author's email: t.p.dale@keele.ac.uk

Chronic Obstructive Pulmonary Disease (COPD) presents a significant, incurable, worldwide health burden, identifying it as a prospective regenerative medicine target. Sourcing cells for lung therapies is challenging; progress has been made in culturing upper airway basal cells but information on the culture of distal cells remains scarce.

Distal lung from human donors was minced and digested. Cells were plated on type I collagen-coated flasks in cFAD medium with rock inhibitor (cFAD+)[1]. Cells were characterised by immunocytochemistry for vimentin, smooth muscle actin, pan-cytokeratin, TP63, cytokeratin-5, E-cadherin, Club cell secretory protein (CCSP), mucin-5AC and β-IV-tubulin. Differentiated using air-liquid interface culture, with measurement of trans-epithelial electrical resistance (TEER), in a matrigel culture system, and seeded to porous collagen scaffolds.

Culture in cFAD+ resulted in predominantly epithelial cells expressing pan-cytokeratin and E-cadherin, TP63 and cytokeratin-5 suggesting a distal airway stem cell (DASC) identity. At air-liquid interface cells developed tight junctions with increased TEER (≥350 Ω.cm2), increased levels of CCSP, stained positively for β-IV-tubulin and mucin-5AC and had visible, motile cilia. Matrigel culture resulted in self-organising, mucus producing organoids, with visible motile cilia on the surface in addition to thin-walled, cavitated, alveolar-like organoids. The DASCs attached readily to collagen scaffolds, had excellent viability and exhibited proliferation on the scaffolds over a 14 day period.

We have successfully isolated and expanded epithelial progenitors from the distal lung tissue of COPD and healthy donors. Preliminary work demonstrates the DASC multipotential differentiation capacity in a variety of formats making them a valuable source for regenerative medicine therapeutic approaches.

Keywords: Disease models; Distal airway stem cells; COPD

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Abstract 1610

HUMAN GROWTH FACTOR FUNCTIONALIZED PHOTO-CLICKABLE HYALURONAN HYDROGELS FOR STEM CELLS 3D CULTURE

Rui J Almeida[1], Marco Esteves[1], Pedro Lavrador[1], Vítor M Gaspar[1], João F. Mano[1]

[1]*University of Aveiro*

Corresponding author's email: ruijorgealmeida@ua.pt

The biofunctionalization of ECM-mimetic polysaccharide hydrogels with human biomolecule cocktails holds great promise for engineering cell-rich living platforms with improved bioactivity and widespread applicability in numerous biomedical applications. To generate such platforms, herein norbornene-functionalized hyaluronan (HA-Nor) and human-based platelet lysates (hPLs) we combined for engineering click chemistry, orthogonally crosslinkable hydrogel platforms that support mesenchymal stromal/stem cells adhesion and bioactivity. Hyaluronan-norbornene functionalization was performed via a straightforward two-pot grafting-to approach and enabled a rapid combination with hPLs and stem cells under mild conditions, giving rise photo-induced thiol-norbornene crosslinkable networks. Hydrogels fabrication with different concentrations of hPLs impacted constructs viscoelastic properties, swelling kinetics and stem cells metabolic activity. Interestingly, cell viability analysis, via live/dead and metabolic assays, demonstrated that 10% w/v PLs in hydrogels promoted higher cell than their non-functionalized counterparts. Moreover, such platforms were also amenable for MSCs 3D spheroids encapsulation. Overall, the synthesized hybrid platform holds great potential for biomedical applications that can benefit from in situ cells and hPLs delivery, particularly in the context of regenerative medicine.

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Abstract 1611

GRAPHENE OXIDE-DOPED GELLAN GUM-PEGDA HYDROGEL MIMICKING THE MECHANICAL AND LUBRICATION PROPERTIES OF ARTICULAR CARTILAGE

Diego Trucco[1,2,3], Lorenzo Vannozzi[1,2], Eti Teblum[4,5], Madina Telkhozhayeva[4,5], Gilbert Nessim[4,5], Saverio Affatato[6], Hind Al-Haddad[1,2], Gina Lisignoli[3], Leonardo Ricotti[1,2]

[1]*The BioRobotics Institute, Scuola Superiore Sant'Anna, Piazza Martiri della Libertà 33, 56127 Pisa, Italy*
[2]*Department of Excellence in Robotics & AI, Scuola Superiore Sant'Anna, Piazza Martiri della Libertà 33, 56127 Pisa, Italy*
[3]*IRCSS Istituto Ortopedico Rizzoli, SC Laboratorio di Immunoreumatologia e Rigenerazione Tissutale, Via di Barbiano, 1/10, Bologna 40136, Italy*
[4]*Department of Chemistry, Bar-Ilan University, Ramat Gan 52900, Israel*
[5]*Bar Ilan Institute for Nanotechnology and Advanced Materials (BINA), Bar-Ilan University, Ramat Gan 52900, Israel*
[6]*IRCSS Istituto Ortopedico Rizzoli, Laboratorio Tecnologie Biomediche, Via di Barbiano, 1/10, Bologna 40136, Italy*

Corresponding author's email: d.trucco@santannapisa.it

Introduction: Articular cartilage (AC) is a specialized connective tissue which provides a low-friction gliding surface, supporting shock-absorption and wear-resistance. Nowadays, conventional strategies show several limitations in restoring chondral defects. This work reports the fabrication of a bilayered structure made of gellan gum (GG) and poly(ethylene-glycol) diacrylate (PEGDA), mimicking mechanical and lubrication of AC in deep and superficial zones. Graphene oxide (GO) was analyzed as lubricant agent.

Methods: Blends of GG and PEGDA were crosslinked by UV-light and magnesium chloride. GO was synthesized following modified Hummer's method1, and embedded into the superficial layer. Wear tests, performed following ISO14243, were performed on a knee simulator. Cytotoxic effects on chondrocytes were assessed by Live/Dead and MTT assays.

Results: Mechanical tests allowed to determine the optimal crosslinking parameters, by combining photo (5 min) and ionic crosslinking with MgCl2, to target the Young's modulus of superficial and deep zone2. The presence of GO into the superficial layer provided a lower coefficient of friction in the kinetic regime (~ 0.03) than the non-doped hydrogels. The wear test confirmed the resistance of the bilayered hydrogel up to 100,000 cycles. The hydrogel formulations did not show any sign of cytotoxicity.

Conclusions: These results are promising in view of the fabrication of a multi-layered synthetic implant for the restoration of AC.

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Keywords: Cartilage substitute; Cartilage mechanical properties; Cartilage lubrication properties

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Abstract 1612

TOWARDS BIOHYBRID LUNG DEVELOPMENT: DE NOVO SYNTHESIS OF ENDOTHELIAL EXTRACELLULAR MATRIX SUPPORTS MONOLAYER STABILITY UNDER FLOW CONDITIONS

Michael Pflaum[1], Katherina Katsirntaki[1], Sophie Jurmann[1], Claudia Hartl[1], Zlata Vukadinovic-Nikolic[1], Axel Haverich[1,2], Bettina Wiegmann[1,2]

[1]*Hannover Medical School*
[2]*German Center for Lung Research*

Corresponding author's email: pflaum.michael@mh-hannover.de