GermanStemCellNetwork

13th GSCN Conference

Speakers & Abstracts

15 – 17 October 2025

Helmholtz Munich

Conference Center Neuherberg







www.gscn.org

Academic Supporter

HELMHOLTZ MUNICH

Funded by DFG Deutsche Forschungsgemeinschaft

BIH Berlin Institute of Health @Charité

TACTiC

DZL Deutsches Zentrum für Lungenforschung

BREATH

Gold Supporter



Silver Supporter



ACTIVE BIOSCIENCE
PROTEINS · CELLS · ANTIBODIES







Bronze Supporter



























Academic Supporter











13th GSCN Conference
15 – 17 October 2025
Helmholtz Munich

Organizer:

German Stem Cell Network (GSCN)

c/o Charité Universitätsmedizin

Luisenstr. 65, 10117 Berlin

GermanStemCellNetwork

Web: www.gscn.org | E-mail: office@gscn.org | Phone: +49 30 450 54 36 38

Dear colleagues and friends, partners and participants,



It is a great pleasure and honor to welcome you all to this 13th German Stem Cell Network Conference in Munich – a city renowned not only for its cultural richness and scientific tradition, but also for its spirit of innovation in modern biomedical research at Helmholtz Munich, Max Planck Society, Fraunhofer Institute as well as the Technical and Ludwig-Maximilian Universities and clinical hospitals. I am delighted to welcome so many outstanding stem cell scientists, clinicians, and representatives from industry, and society gathering at the brand new conference center at Helmholtz Munich in Neuherberg, in the North of Munich. Your commitment to participate, your curiosity, and courage to push boundaries, will generate an excellent atmosphere for an exciting meeting over the three days gathering from 15 till 17 October 2025.

I am looking forward to over 400 participants who will come to discuss stem cell research in all flavors and debate new technologies, first clinical study results and fascinating data, shown on many posters and in oral presentations from junior and senior colleagues, keynote speakers and award lectures. An interesting and broad industry exhibition will offer latest technologies and personal contact in combination with industry sessions of GSCN industry partners. There will also be the non-PI meeting, as well as GSCN mentoring program workshop. Finally, we will enjoy two social events to continue networking after working hours. Check out the whole program on our online platform (www.gscn-conferences.org -> Program & Speakers 2025).

Stem cell research today represents both immense promise and great responsibility. It offers hope for new therapies for currently incurable diseases, deeper insights into human development, and challenges us to reconsider ethical, societal, and scientific questions in entirely new ways.

At this point, I would like to extend my sincere thanks to everyone who made this conference possible: the GSCN office, the local organizers and program committee, the speakers, the Helmholtz Munich staff who made this happen, and of course, all of you — whose presence and contributions will make this event a vibrant space for knowledge, inspiration, and connection.

I wish all of us stimulating discussions, meaningful encounters, and fresh ideas to carry back to our labs, clinics, and communities.

Thank you very much — and let's have a successful conference!

Heiko Lickert
GSCN President

For the program committee

Sina Bartfeld (Berlin) · *Daniel Besser* (Berlin) · *Magdalena Götz* (Munich) · *Matthias Hebrok* (Munich) · *Karl-Ludwig Laugwitz* (Munich) · *Heiko Lickert* (Munich) · *Thomas Schwarz-Romond* (Munich) · *Fabian Theis* (Munich) · *Claudia Waskow* (Jena)

Table of contents

Conference Information 5

GSCN Awards 6

ELSA Symposium 6

Social Events 7

Floor plan industry exhibition 8

Floor plan Build23 10

Helmholtz Campus site plan 11

Program Overview 12

Public outreach event 15

Program 16

Speaker abstracts ..... **28 – 129**

Keynote lectures 30

Presidential Symposium lectures 36

Selected presentations 41

Strategic sessions presentations 100

Company presentations 111

Poster abstracts ..... **130 – 291**

Basic, translational & applied hematopoiesis (P001 – P006) 132

Pluripotency, differentiation & PSC-derived models (P007 – P047) 139

Stem cells in aging & disease (cancer stem cells) (P048 – P051) 183

Tissue-derived stem cells & organoids (P052 – P072) 188

Computational stem cell biology & systems biology (P073 – P081) 212

Stem cells in disease modeling & drug development (P082 – P124) 222

Stem cells in regenerative therapies (P125 – P144) 269

Author Index 292

List of Participants 296

Supporters & Exhibitors 316

Conference Information

Venue

Helmholtz Munich
Conference Center (Building 3522)
Ingolstädter Landstr. 1
85764 Neuherberg

Date

Wednesday, 15 October – Friday, 17 October 2025

Internet

Internet access via Wireless LAN is free of charge.

Registration

Regular fee	€ 620,-
Regular member fee	€ 400,-
Student fee	€ 310,-
Student member fee	€ 200,-

The registration fee includes admission to all scientific sessions, additional scientific events, poster and commercial exhibition, the get-together on 15 October and the networking evening on 16 October, lunch and coffee breaks, free internet access, conference bag including badge and access to the GSCN web app.

Poster Exhibition

Posters will be displayed during the conference in two sessions in Build23. Authors are asked to be present at their poster during the poster session. Even numbers will be presented on Wednesday, 15 October 2025, 17:30 – 18:30 h and odd numbers will be presented 18:30 – 19:30 h. On Thursday, 16 October 2025 even numbers will be presented from 16:30 – 17:30 h and odd numbers from 17:30 – 18:30 h. You will find the number of your poster in this abstract volume. Please prepaire the size DIN A0 portrait for the poster. Otherwise, there are no specifications. Posters in poster session I should be mounted on Wednesday 12:00 – 16:30 h and removed latest on Thursday, 12:00 h. Posters in Poster Session II should be mounted on Thursday 12:00 – 16:30 h and should be removed latest on Friday at 14:00 h.

Poster Session I

Wednesday, 15 October 2025, 17:30 – 19:30 h

Basic, translational & applied hematopoiesis: P001 - P006
Pluripotency, differentiation & PSC-derived models: P007 - P047
Stem cells in aging & disease (cancer stem cells): P048 - P051
Tissue-derived stem cells & organoids: P052 - P072



The poster session I is supported by Biomerieux

Poster Session II

Thursday, 16 October 2025, 16:30 – 18:30 h

Computational stem cell biology: P073 - P081
Stem cells in disease modeling & drug dev.: P082 - P124
Stem cells in regenerative therapies: P125 - P144



The poster session II is supported by Miltenyi

GSCN Awards

Scientific awards 2025

A top-class commission supported the GSCN in choosing this year´s awardees. The three awardees will give a presentation in the Presidential Symposium on Thursday, 16 October 2025 at 14:00 - 16:15 h.

GSCN 2025 Young Investigator Award:	***Aydan Bulut-Karslioglu***, Max Planck Institute for Molecular Genetics, Berlin
GSCN 2025 Hilde Mangold Award:	**Michaela Frye**, German Cancer Research Center, Heidelberg
GSCN 2025 Publication of the Year Award:	***Moritz Mall***, German Cancer Research Center, Heidelberg and ***Bryce Lim***, SZ, London, UK

Travel awards

The following participants have been selected for the GSCN travel awards:
– ***Annasara Artioli***, Mannheim
– ***Mari Ozaki***, Dublin, IE
– ***Meiying Xue***, Berlin
– ***Sohana Islam Khan***, Halle
– ***Susanna Quach***, Berlin

Poster awards

There will be two poster awards for each poster session. Members of the poster jury have a red dot on their badge – please present them your poster with due attention. The awardees will receive € 500 each. Authors are asked to be present at the poster award ceremony on Friday, 17 October 2025 at 15:30 – 15:50 h in the plenary hall.

The poster awards are supported by Active Biosciense + PeptiGrowth



Elsa Symposium

Embryo models meet genome engineering: A dialog on ethics and innovation

Recent advances in three-dimensional tissue culture and matrix interactions have led to the development of increasingly authentic models of human physiology, enabling researchers to study inherited and acquired diseases. This trend is evident in various somatic organoids, including neuro-organoids, and has also been applied to embryo models, which are now reaching advanced developmental stages. Genome editing tools have been widely used in organoid research to interfere with disease-associated pathways or repair inborn genetic mutations, raising only few ethical concerns. However, when applied to human embryo models, these techniques spark more intense ethical discussions. At this year's conference, we aim to initiate a dialog on these critical issues and are honored to welcome Jacob Hanna, Weizmann Institute of Science, Rehovot, Israel, as our keynote speaker for Wednesday evening. During the ELSA Symposium on Thursday morning, Dr. Hanna will join us to explore the latest advancements in the rapidly growing field of embryo models. We will also examine the emerging concept of human-on-a-chip models, which serve as in vitro avatars for patient-specific studies. This raises fundamental questions about the characteristics that render in vitro models entities with some form of human moral status. While this symposium will not provide comprehensive answers to these complex questions, we expect it to be a thought-provoking starting point for a broader debate on the ethics and implications of embryo modeling and genome engineering.

Social Events

Get-together

On Wednesday, 15 October 2025, from 20:30 – 22:00 h, we invite all participants and exhibitors to a get-together with an evening buffet in the mensa, which is just a 2-minute walk from the conference center. Starting at 22:00 h, buses will be available to transfer to the Garching-Hochbrück subway station (U6 towards Garching and Munich City Center), which is located in the immediate vicinity of most hotels in Garching. Bus departure points can be found on the map on page 10.

The Get-together is supported by
FUJIFILM Wako Chemicals Europe GmbH

Networking Event

The GSCN Networking Event with dinner buffet and DJ is open for all participants and exhibitors and will take place on Thursday, 16 October 2025, 19:00 – 01:00 h at the Gasthof Neuwirt in Garching. From 18:30 h, there will be a bus transfer from the conference center to Gasthof Neuwirt. The departure point for the buses can be found on the map on page 10. After the event, all participants can take the subway (U6) from the Garching subway station which is located directly behind the Gasthof Neuwirt. It is only one station to Garching-Hochbrück.

Adress:
Gasthof Neuwirt Garching, Münchener Str. 10, 85749 Garching

The Networking Event is supported by
STEMCELL Technologies GmbH



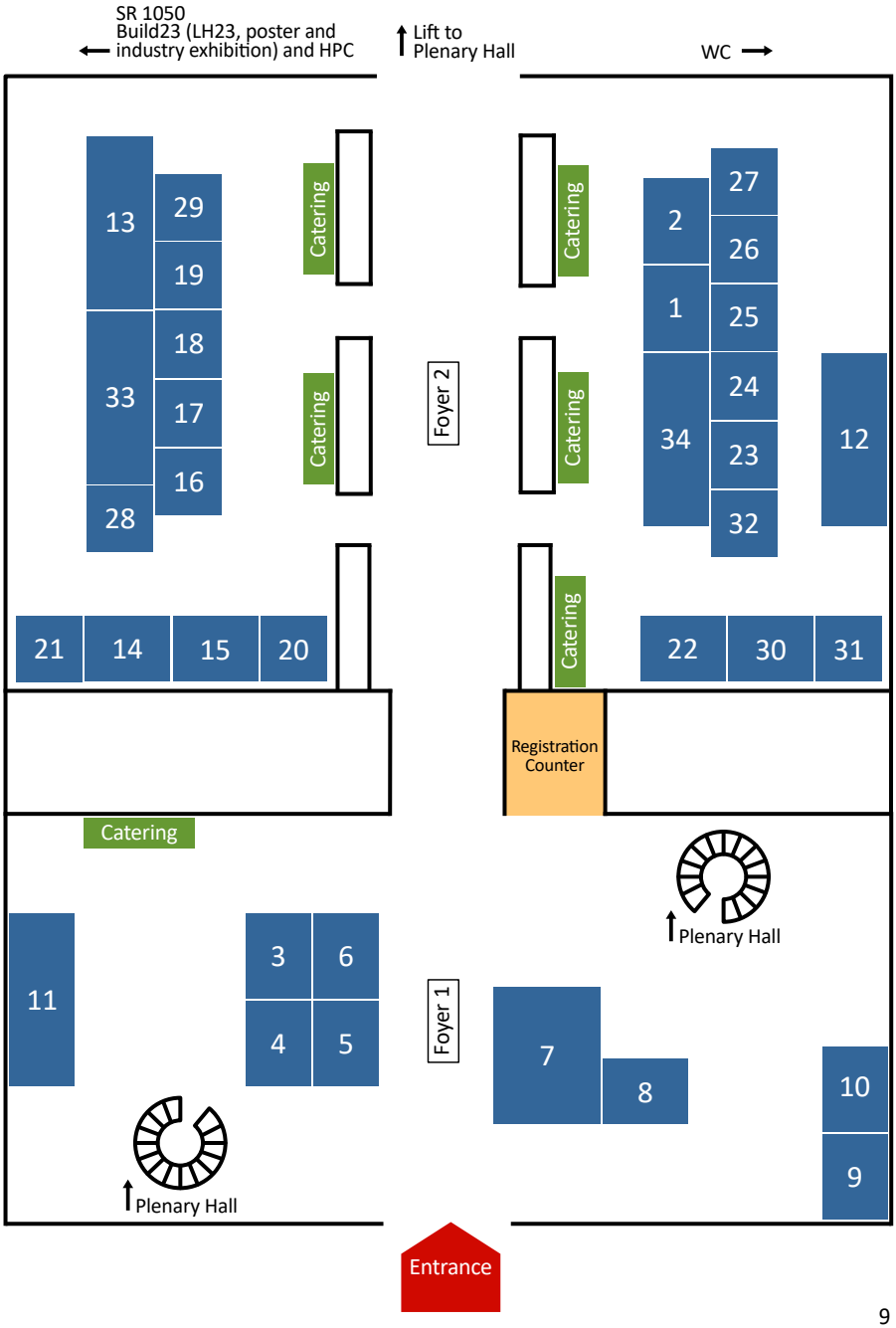
List of exhibitors (grey ones are located in Build23)

Sorted in alphabetical order COMPANY	NUMBER
Active Bioscience and PeptiGrowth*	5
Amsbio*	18
Becton Dickinson (BD)*	34
BioLamina*	14
Biomerieux Deutschland GmbH*	10
Biomol GmbH*	17
Bio-Techne GmbH*	36
Biozym Scientific GmbH	25
Cellseeker*	20
Cenibra GmbH	19
Eppendorf SE*	4
Evotec*	8
faCellitate	24
FUJIFILM Wako Chemicals Europe GmbH*	37
ibidi GmbH	29
Integrated DNA Technologies*	30
iotaSciences*	3
MACHEREY-NAGEL GmbH & Co. KG*	26
MedChemExpress	27
Miltenyi Biotec B.V. & Co. KG*	1
Minaris*	9
New England Biolabs	12
Nippon Genetics Europe*	16
Novogene Europe	32
OMNI Life Science GmbH & Co. KG*	35
PELOBiotech GmbH*	13
Proteintech	33
Qkine*	22
SARSTEDT AG & Co. KG	28
Sartorius*	6
Stem Genomics*	2
STEMCELL Technologies GmbH*	7
SYNENTEC GmbH	11
Telight	31
Thermo Fisher Scientific*	15
VectorBuilder*	38
Xceltis GmbH	21
Zymo Research Europe	23

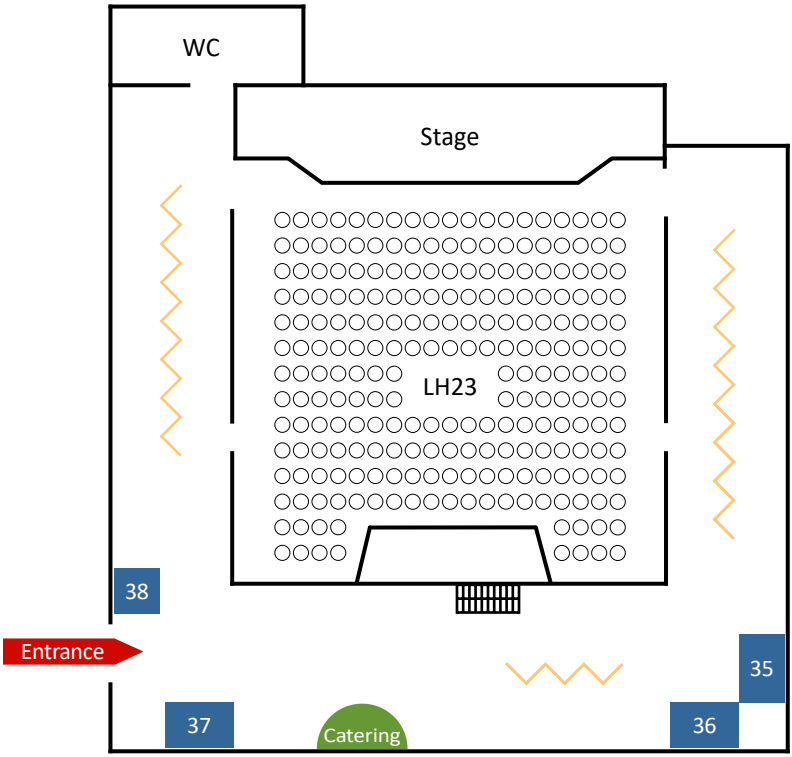
Sorted by booth number NUMBER	COMPANY
1	Miltenyi Biotec B.V. & Co. KG*
2	Stem Genomics*
3	iotaSciences*
4	Eppendorf SE*
5	Active Bioscience and PeptiGrowth*
6	Sartorius*
7	STEMCELL Technologies GmbH*
8	Evotec*
9	Minaris*
10	Biomerieux Deutschland GmbH*
11	SYNENTEC GmbH
12	New England Biolabs
13	PELOBiotech GmbH*
14	BioLamina*
15	Thermo Fisher Scientific*
16	Nippon Genetics Europe*
17	Biomol GmbH*
18	Amsbio*
19	Cenibra GmbH
20	Cellseeker*
21	Xceltis GmbH
22	Qkine*
23	Zymo Research Europe
24	faCellitate
25	Biozym Scientific GmbH
26	MACHEREY-NAGEL GmbH & Co. KG*
27	MedChemExpress
28	SARSTEDT AG & Co. KG
29	ibidi GmbH
30	Integrated DNA Technologies*
31	Telight
32	Novogene Europe
33	Proteintech
34	Becton Dickinson (BD)*
35	OMNI Life Science GmbH & Co. KG*
36	Bio-Techne GmbH*
37	FUJIFILM Wako Chemicals Europe GmbH*
38	VectorBuilder*

**Partners of the GSCN Quiz. Win one of three Amazon 200 € Gift Vouchers.*

Floor plan industry exhibition



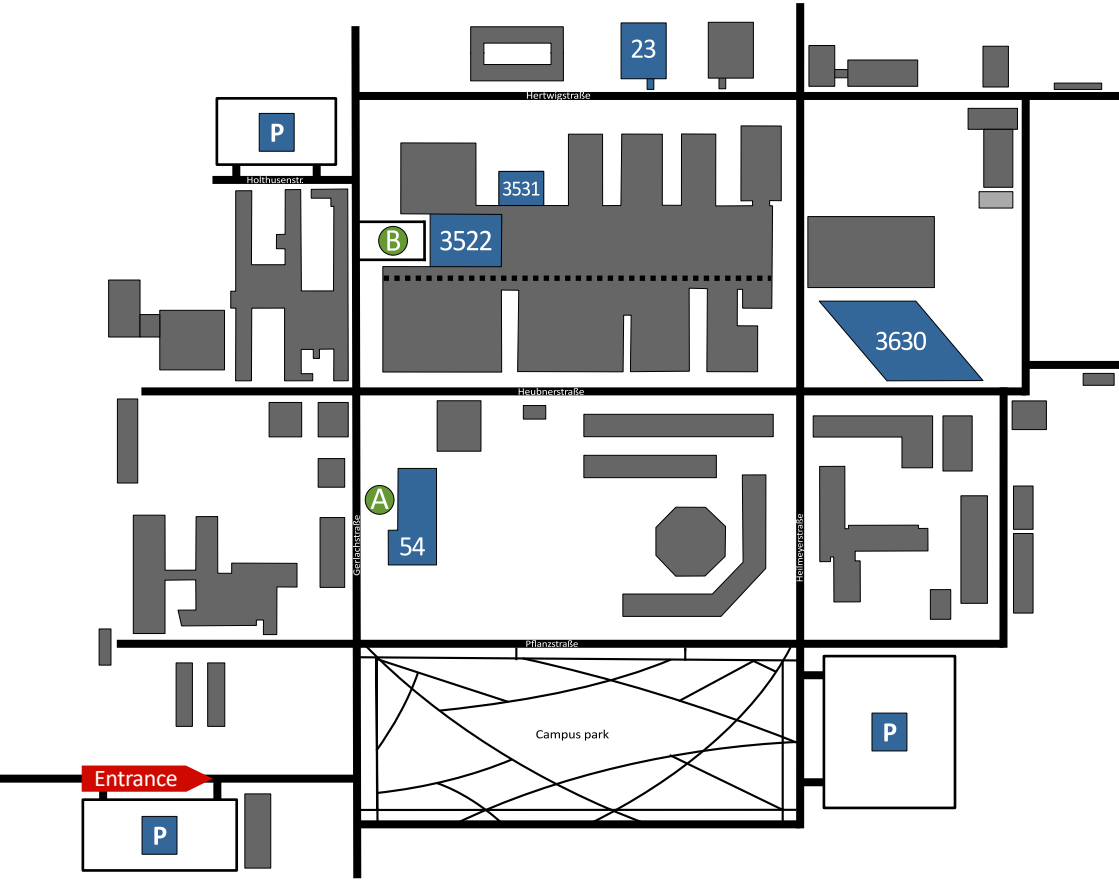
Floor plan Build23 – Poster and industry exhibition



Number	Company
35	OMNI Life Science GmbH & Co. KG*
36	Bio-Techne GmbH*
37	FUJIFILM Wako Chemicals Europe GmbH*
38	VectorBuilder*

**Partners of the GSCN Quiz. Win one of three Amazon 200 € Gift Vouchers.*

Helmholtz Munich campus site plan



23	Build23 (LH23)
54	Mensa
3522	Conference Center
3531	Seminar room 1050
3630	Helmholtz Pioneer Campus (HPC)
A	Bus transfer to the subway station Garching-Hochbrück. Wed., 15 October, 22:00 h
B	Bus to the networking event, Thu., 16 October, 18:30 h

Program Overview Wednesday

	Helmholtz Munich Conference Center				
09:00 – 19:00	Registration				
	Plenary Hall	Seminar room 1050	LH23	Seminar room HPC*	Foyer Build23
11:00 – 11:30	Opening				
11:30 – 12:15	**Keynote lecture I** *Viviane Tabar*				
12:15 – 13:00	**Keynote lecture II** *Timm Schröder*				
13:00 – 14:30	Lunch break (Mensa) / Industry exhibition				
14:30 – 16:00	**Concurrent scientific working group session I**				
	Stem cells in regenerative therapies	Tissue-derived stem cells & organoids	Stem cells in disease modelling & drug development		
16:00 – 16:30	Break time / Industry exhibition (CC & Build23)				
16:30 – 17:30			GSCN Members Meeting		
17:30 – 18:30					**Poster session I** even numbers
18:30 – 19:30					**Poster session I** uneven numbers
19:45 – 20:30	**Keynote lecture III** (on ELSA topics) *Jacob Hanna*				
20:30 – 22:00	Informal GSCN get-together with dinner buffet at the Mensa				

**Helmholtz Pioneer Campus*

Program Overview Thursday

	Helmholtz Munich Conference Center				
08:00 – 18:00	Registration				
	Plenary Hall	Seminar room 1050	LH23	Seminar room HPC*	HPC* – VIP
09:00 – 10:30	**Concurrent scientific working group session II**				
	Basic, translational and applied hematopoiesis	TACTIC Symposium on iPSC-based heart Repair	Pluripotency, differentiation & PSC-derived models	Stem cells in disease modeling & drug development II	Elsa Symposium
10:30 – 11:00	Break time / Industry exhibition (CC & Build23)				
11:00 – 12:35 SR1050 until 12:45 LH23 &HPC until 12:25	**Industry session: "Technologies from GSCN industry partners"**				
	Eppendorf SE STEMCELL Technologies GmbH Active Bioscience/ PeptiGrowth	Thermo Fischer Scientific Biomerieux DE GmbH Miltenyi Biotec B.V. & Co. KG BioLamina Sartorius	iotaSciences Integrated DNA Technologies Evotec Stem Genomics	Qkine Minaris OLS OMNI Life Science Bio-Techne	
12:30 – 14:00	Lunch break (Mensa) / Industry exhibition				
14:00 – 14:45	Presidential Invite: *Wolfram-Hubertus Zimmermann*	**Presidential Symposium**			
14:45 – 15:15	Young Investigator Award: *Aydan Bulut-Karslioglu*				
15:15 – 15:45	Hilde Mangold Award: *Michaela Frye*				
15:45 – 16:15	Publication of the Year Award: *Moritz Mall, Bryce Lim*				Foyer Build23
16:15 – 16:30	Break time / Industry exhibition (CC & Build23)				
16:30 – 17:30					**Poster session II** even numbers
17:30 – 18:30					**Poster session II** uneven numbers
19:00 – 01:00	GSCN networking event with dinner buffet & DJ at Gasthof Neuwirt, Garching				

**Helmholtz Pioneer Campus*

Program Overview Friday

	Helmholtz Munich Conference Center			
08:00 – 16:00	Registration			
	Plenary Hall	Seminar room 1050	LH23	Seminar room HPC*
09:00 – 10:30	**Concurrent scientific working group session III**			
	Stem cells in aging & disease	DZL/Breath symposium: stem cells in lung research	Pluripotency, differentiation & PSC-derived models II	Computational stem cell biology
10:30 – 11:00	Break time / Industry exhibition (CC & Build23)			
11:00 – 12:30	**Concurrent strategic working group session**			
	Technologies in stem cell research	Career development	Clinical trials & regulatory affairs	
12:30 – 14:00	Lunch break (Mensa) / Industry exhibition			
14:00 – 14:45	**Keynote lecture IV** *Hongkui Deng*			
14:45 – 15:35	**Keynote lecture V** *Maria-Elena Torres-Padilla*			
15:35 – 16:00	Poster Award Ceremony and closing remarks			
16:00	END OF CONFERENCE			

GSCN public outreach event (in German) with panelists
at Presseclub München, Marienplatz 22, 80331 München

Stammzellforschung bei Diabetes, Alterung und seltenen Erkrankungen.
Chancen, Risiken und Perspektiven

**Helmholtz Pioneer Campus*

Public outreach event (in German)

Stammzellforschung bei Diabetes, Alterung und seltene Erkrankungen. Chancen und Risiken

Wann: **Freitag, 17. Oktober 2025, 18:30 – 20:00 Uhr**

Wo: Presseclub München, Marienplatz 22, 80331 München

Programm: 18:30 – 20:00 Uhr: Podiumsdiskussion & Kurzfilme

20:00 – 20:30 Uhr: Ausklang mit Gesprächen & Getränken

Auf dem Podium

Prof. Dr. Anette-Gabriele Ziegler, Helmholtz Munich, Expertin für Diabetes

Prof. Dr. Heiko Lickert, Helmholtz Munich, Experte für Diabetes & Stammzellen

Dr. Ole Pless, Frauenhofer ITMP Hamburg, Experte für Wirkstofftestung und seltene Erkrankungen

Prof. Dr. Hartmut Geiger, Universität Ulm, Experte für Alterung

Moderation: *Dr. Stefanie Seltmann*, Wissenschaftsjournalistin, Heidelberg

Beschreibung

Haare wachsen ein Leben lang. Wunden heilen wieder. Zellen im Blut erneuern sich alle drei Monate. Verantwortlich für diese Phänomene sind Stammzellen, eine ganz besondere Zellart in unserem Körper: Ein Leben lang können sie sich teilen, vermehren und in alle verschiedenen Körperzellen entwickeln. Wenn bei diesen fundamentalen Vorgängen Fehler passieren, können Krankheiten entstehen. Daher liefert die Erforschung von Stammzellen wichtige Erkenntnisse für die Medizin. Perspektiven für Zelltherapien und Testsysteme entstehen. Unsere Themen:

> Wie weit sind Zelltherapien bei Diabetes? Wie früh kann Diabetes überhaupt festgestellt werden?
>
> Stammzellen als Detektiv? Wie findet man das richtige Medikament für eine seltene Erkrankung?
>
> Stammzellen als Jungbrunnen? Alle altern – können Stammzellen beim gesunden Altern helfen?

Bei der Podiumsdiskussion berichten die eingeladenen Wissenschaftler*innen, welche Rolle Stammzellen bei der Erforschung von Diabetes, seltenen Erkrankungen und dem Alterungsprozess spielen. Kurzfilme stellen die Forschung der Wissenschaftler*innen vor. In der Diskussion dreht es sich um Stand und Zukunft der Stammzellforschung sowie neue Behandlungsmöglichkeiten. Im Anschluss an die Veranstaltung sind alle herzlich eingeladen, das Gespräch bei Getränken fortzuführen.

Program Wednesday, 15 October 2025

11:00 - 11:30 Plenary Hall	**Opening GSCN Conference 2025**
11:00 - 11:15	Welcome Helmholtz Munich *Martin Hrabě de Angelis*, Research Director Hemholtz Munich
11:15 - 11:30	Opening of GSCN Conference 2025 *Heiko Lickert*, President GSCN
11:30 - 12:15 Plenary Hall	**Keynote lecture I** supported by FUJIFILM Wako Chemicals Europe K1 - Advances in cell therapy for Parkinson's disease: Insights from the results of a phase 1 trial and future directions *Viviane Tabar*, Memorial Sloan Kettering Cancer Center, New York, USA Chair: *Magdalena Götz*
12:15 - 13:00 Plenary Hall	**Keynote lecture II** K2 - Long-term single live cell quantification: New tools for old questions *Timm Schroeder*, Biosystems Science and Engineering (BSEE), ETH Zürich, Basel, Switzerland Chair: *Claudia Waskow*
13:00 - 14:30 Mensa	Lunch break / Industry exhibition
14:30 - 16:00	**Concurrent scientific working group session I**
14:30 - 16:00 Plenary Hall	**Stem cells in regenerative therapies** supported by Eppendorf Chairs: *Ulrich Martin* & *Wolfgang Wagner*
14:30 - 15:00	T01 - Therapeutic CAR-T cell products: What can we learn for pluripotent stem cell based products? *Ulrike Köhl*, Fraunhofer IZI, Leipzig
15:00 - 15:15	T02 - Maximizing gene editing efficiency for enhanced clinical-grade iPSCs via sequential factor delivery *Thomas Berger*, Catalent, Langenfeld
15:15 - 15:30	T03 - Off the shelf immune-shielded stem cell-derived macrophages for clinical applications *Stefanie Pfaender*, Evotec, Cell Therapy, Göttingen
15:30 - 15:45	T04 - Prototypical ten-week end-to-end process for autologous Parkinson's disease cell therapy *Diana Haberhausen*, Mitenyi Biotec, Bergisch Gladbach

15:45 - 16:00 T05 - Enhancing stem cell derived beta cell survival in hypoxia and nutrient deprivation with small molecules
Väinö Lithovius, Helmholtz Diabetes Center, Munich

14:30 - 16:00 **Tissue-derived stem cells & organoids** supported by Sartorius
SR1050 Chairs: *Sina Bartfeld* & *Anne Grapin-Botton*

14:30 - 15:00 T06 - Mapping the transcriptome and DNA-methylome at the single cell level decodes stemness across the young and injured brain
Ana Martin-Villalba, DKFZ Heidelberg

15:00 - 15:15 T07 - Manipulation of global protein translation rates and stem cell self-renewal by CRISPR mediated activation of rRNA transcription
Stefan Stricker, LMU & Helmholtz Munich

15:15 - 15:30 T08 - Slit–Robo signaling controls early liver zonation during fetal development
Dominic Grün, Würzburg Institute of Systems Immunology

15:30 - 15:45 T09 - Unveiling human cardiac regenerative mechanisms in iPSC-derived epicardioids
Tobias Ellinger, TUM Munich

15:45 - 16:00 T10 - Human-specific morphoregulatory signatures in basal radial glia characterize neocortex evolution
Mareike Albert, TU Dresden

14:30 - 16:00 **Stem cells in disease modeling & drug development**
LH23 Chairs: *Oliver Brüstle* & *Harald Stachelscheid* supported by Evotec SE

14:30 - 15:00 T11 - Epicardioids: Bridging cardiac development with disease and therapy
Alessandra Moretti, TUM School of Medicine and Health, Munich

15:00 - 15:15 T12 - AI-guided protein design of synthetic growth factor mimetics
Thomas Schlichthärle, TUM School of Natural Science, Munich

15:15 - 15:30 T13 - Enhanced fibrosis modeling in human skin organoids through hiPSC-derived macrophage integration
Amelie Reigl, Fraunhofer Institute for Silicate Research (ISC), Würzburg

15:30 - 15:45 T14 - Heart organoids and engineered heart tissue as hiPSC-derived in vitro models for Fabry cardiomyopathy
Anna Reinelt, University hospital Eppendorf (UKE), Hamburg

15:45 - 16:00	T15 - iPSC-driven drug discovery identifies sildenafil as a drug candidate for the mitochondrial disease Leigh syndrome *Annika Zink*, University hospital Düsseldorf
16:00 - 16:30 CC & Build23	Break time / Industry exhibition
16:30 - 17:30 LH23	**General Member Meeting** Executive Board & Managing Director *Heiko Lickert*, *Claudia Waskow*, *Sina Bartfeld*, *Michael Cross*, *Daniel Besser* (excused Andreas Trumpp)
17:30 - 19:30 Build23	**Poster session I - Poster number P001 - P072** supported by Biomerieux (with beverages) Basic, translational & applied hematopoiesis: P001 - P006 Pluripotency, differentiation & PSC-derived models: P007 - P047 Stem cells in aging & disease (cancer stem cells): P048 - P051 Tissue-derived stem cells & organoids: P052 - P072 Even numbers: 17:30 - 18:30 Odd numbers: 18:30 - 19:30
19:45 -20:30 Plenary Hall	**Keynote lecture III (on ELSA topics)** K3 - Synthetic ex utero embryogenesis: From naive pluripotent stem cells to bona fide embryo models *Jacob Hanna*, Weizmann Institute of Science, Rehovot, Israel Chair: *Tobias Cantz*
20:30 - 22:00 Mensa	**GSCN Get-together for all participants** supported by FUJIFILM Wako Chemicals **Dinner buffet**

Program Thursday, 16 October 2025

09:00 - 10:30	**Concurrent scientific working group session II**
09:00 - 10:30 Plenary Hall	**Basic, translational & applied hematopoiesis** supported by Thermo Fisher Chairs: *Nina Cabezas-Wallscheid* & *Michael Milsom*
09:00 - 09:30	T16 - Blood stem cell expansion: Technology to biology *Adam Wilkinson*, Cambridge Stem Cell Institute, UK
09:30 - 09:45	T17 - C57BL/6 Kit-mutant immunodeficient mouse models uncover mechanism of successful hematopoietic stem cell xenotransplantation *Julia Fröbel*, Leibniz Institute on Aging – FLI, Jena
09:45 - 10:00 Plenary Hall	T18 - From codons to blood: A new perspective on hematopoietic differentiation *Marius Külp*, Goethe University, Frankfurt

10:00 - 10:15 T19 - Multimodal omics identify an age resilient multipotent progenitor population that preserves youthful identity and function in the hematopoietic system
Mari Carmen Romero-Mulero, ETH Zurich, CH

10:15 - 10:30 T20 - Development of accessible ex vivo culture models of the human hematopoietic bone marrow niche
Sigrid Uxa, University Leipzig

09:00 - 10:30 SR1050 **TACTIC symposium on iPSC-based heart repair**
BMBF-funded project "Tailored Application for individualized Cell Therapy with iPSC-derived Cardiomyocytes (TACTiC)"
Chairs: *Ulrich Martin* & *Robert Zweigerdt*

09:00 - 09:30 T21 - Deep immune-phenotyping of HLA-homozygous iPS-cardiomyocytes
Dirk Strunk, PMU Salzburg, Austria

09:30 - 10:00 T22 - Biological-engineering and application of a myocardial patch
Joost Sluijter, UMC Utrecht, The Netherlands

10:00 - 10:15 T23 - Large-scale production of human pluripotent stem cell–derived cardiomyocytes
Kevin Cyrys, MH Hanover

10:15 - 10:30 T24 - Automated myocardial cell injection
Raphael Mönkemöller, LU Hanover

09:00 - 10:30 LH23 **Pluripotency, differentiation & PSC-derived models**
Chairs: *Mina Gouti* & *Hans Schöler*

09:00 - 09:30 T25 - Developing a safe and immune-evasive stem cell source for off-the-shelf therapeutic products
Andras Nagy, Lunenfeld-Tanenbaum Research Institute, Mount Sinai Hospital, Toronto, Canada

09:30 - 09:45 T26 - Lineage single-cell multi-omics identify novel regulators and barriers in iPSC reprogramming
Angeliki Spathopoulou, University of Innsbruck, Austria

09:45 - 10:00 T27 - You shall not pass - functional pluripotency testing using hiPSCore
Jochen Dogner, Leibniz-Institut für umweltmedizinische Forschung (IUF) Düsseldorf

10:00 - 10:15 T28 - PluripotencyScreen – quantification of early cell fate decisions of iPSC based on DNA methylation
Kira Zeevaert, UK RWTH Aachen

10:15 - 10:30 T29 - Does gene dosage influence cell fate during human gastrulation?
Theresa Gross-Thebing, Gurdon Institute, Cambridge, UK

09:00 - 10:30 HPC	**Stem cells in disease modeling & drug development II** Chairs: *Jan Eickhoff* & *Karl-Ludwig Laugwitz* supported by Bio-Techne
09:00 - 09:30	T30 - Modulating reactive astrocytes in Alzheimer's disease *Bastian Zimmer*, Evotec SE, Hamburg
09:30 - 09:45	T31 - Evaluating the potential of Bruton's tyrosine kinase inhibitors to modulate microglial inflammation to attenuate multiple sclerosis progression *Alica Blenkle*, Fraunhofer ITMP, Göttingen
09:45 - 10:00	T32 - A reproducible human brain tissue model to study physiological and disease-associated microglia phenotypes *Dominik Paquet*, LMU Munich
10:00 - 10:15	T33 - Integration of CNTNAP2-deficient neurons into defined E/I networks unmasks functional pathophenotypes *Jasmin Aicher*, University of Bonn
10:15 - 10:30	T34 - Capturing disease severity in LIS1-lissencephaly reveals proteostasis dysregulation in patient-derived forebrain organoids *Matteo Gasparotto*, ZI Mannheim
09:00 - 10:30 HPC-VIP	**ELSA symposium** Panel: *Jacob Hanna* and *Maria-Elena Torres-Padilla* Moderation: *Tobias Cantz*
10:30 - 11:00 CC & Build23	Break time / Industry exhibition supported by Minaris
11:00 - 12:45	**Industry session: "Technologies from GSCN industry partners"**
11:00 - 12:35 Plenary Hall	**Main Supporter** Chairs: *Magdalena Götz* & *Hartmut Geiger*
11:05 - 11:30	C01 - Tackling leachables in stem cell bioprocessing: Ensuring safety and scalability in single-use systems for clinical studies *Philipp Nold*, Eppendorf SE
11:30 - 12:10	C02 - Advance your research with genetically diverse human induced pluripotent stem cell-derived models and a novel mRNA-lipid nanoparticle platform to rapidly generate functional forebrain neurons *Katharina Debowski*, STEMCELL Technologies GmbH
12:10 -12:35	C03 - Forging the future of regenerative medicine with synthetic peptide growth factors *Robert Brownlee*, Opes Diagnostics
11:00 - 12:45 SR1050	**Supporter II** Chairs: *Hannes Klump* & *Ana Martin-Villalba*

11:05 - 11:25	C04 - Approaches for optimizing pluripotent stem cell spheroid expansion in multi-liter bioreactors through the use of constant medium perfusion *David Kuninger*, Thermo Fisher Scientific
11:25 - 11:45	C05 - The world's fastest PCR-based mycoplasma detection assay *Sabine Hahnau-Jurth*, Biomerieux Deutschland GmbH
11:45 - 12:05	C06 - Enabling reliable PSC culture for advanced research applications *Hannah Johannsen*, Miltenyi Biotech B.V. & Co. KG
12:05 - 12:25	C07 - The role of full-length laminins in enhancing in vitro cellular microenvironments *Chiara Annunziata*, BioLamina
12:25 - 12:45	C08 - Live-cell 3D cell imaging innovations *Marco Benkisser-Petersen*, Sartorius
11:00 - 12:25 LH23	**Supporter III** Chairs: *Thomas Braun* & *Andreas Kurtz*
11:05 - 11:25	C09 - Recent advancements in gentle single-cell handling: The iotaSciences way *Matthias Hoja*, iotaSciences
11:25 - 11:45	C10 - Alt-R™ CRISPR capabilities to accelerate your next breakthrough *Katarzyna Julia Duda*, Integrated DNA Technologies
11:45 - 12:05	C11 - Cell therapy at Evotec – end-to-end from process development to clinical manufacturing *Matthias Austen*, Evotec
12:05 - 12:25	C12 - What we've discovered: Opening Pandora's box of genomic mutations using NGS technology *Reda Zenagui*, Stem Genomics
11:00 - 12:30 HPC	**Supporter IV** Chairs: *Thomas Schwarz-Rommond* & *Zoltan Ivics*
11:05 - 11:25	C13 - Commercial media formulations vs chemically defined home-brewed media for iPSC maintenance *Liz Stewart*, Qkine
11:25 - 11:45	C14 - A global vision for cell and gene therapies *Karin Abitorabi*, Minaris
11:45 - 12:05	C15 - Reliable 3D cell models – high-yield cultivation meets precision sorting *Markus Uhrig*, OLS OMNI Life Science

12:05 - 12:25 C16 - Enhancing reproducibility in stem cell and organoid research: Strategies for consistent culture and characterization
Volker Arps, Bio-Techne

12:30 - 14:00 Lunch break / Industry exhibition supported by IDT Germany GmbH
Mensa

14:00 - 16:15 **Presidential Symposium**
Plenary Hall Chair: *Heiko Lickert* & *Claudia Waskow*

14:00 - 14:45 **Presidential Invite**
PS1 - Translation of tissue engineered heart repair
Wolfram-Hubertus Zimmermann, University Hospital Göttingen

14:45 - 15:15 **GSCN 2025 Young Investigator Award**
PS2 - Molecular regulation of mammalian dormancy
Aydan Bulut-Karslıoğlu, Max Planck Institute of Molecular Genetics, Berlin

15:15 - 15:45 **GSCN 2025 Hilde Mangold Award**
PS3 - Harnessing RNA Modifications for Cancer Therapies
Michaela Frye, DKFZ, Heidelberg

15:45 - 16:15 **GSCN Publication of the Year 2024 - 2025 Award**
PS4 - The role of safeguard repressors in cancer
Bryce Lim, Moritz Mall, DKFZ, Heidelberg

16:15 - 16:30 Break time / Industry exhibition supported by Biolamina
CC & Build23

16:30 - 18:30 **Poster session II - Poster number P073 - P144** supported by Miltenyi
Build23 (with beverages)
Computational stem cell biology: P073 - P081
Stem cells in disease modeling & drug dev.: P082 - P124
Stem cells in regenerative therapies: P125 - P144

Even numbers: 16:30 - 17:30
Odd numbers: 17:30 - 18:30

19:00 - 01:00 **GSCN networking event for all participants** supported by STEMCELL Technologies
The entrance fee is collected on site
Event location: Gasthof Neuwirt, Garching
Münchener Str.10
85748 Garching

Program Friday, 17 October 2025

09:00 - 10:30 **Concurrent scientific working group session III**

09:00 - 10:30 Plenary Hall
Stem cells in aging & disease
Chairs: *Hartmut Geiger* & *Simon Haas*

09:00 - 09:30 T35 - Tracing clonal architecture and differentiation trajectories in cancer using single cell barcoding
Michael Rieger, Goethe University Frankfurt

09:30 - 09:45 T36 - Role of aging and environmental stress in the decline of lung stem cell function and regenerative capacity
Maja Funk, Helmholtz Munich

09:45 - 10:00 T37 - Investigating aging and telomere biology in lung disease using induced pluripotent stem cell derived models
Mari Ozaki, Royal College of Surgeons (RCSI), Dublin, Ireland

10:00 - 10:15 T38 - High fat diet affects the stromal niche signaling in the porcine intestine
Falk Farkas, Helmholtz Munich

10:15 - 10:30 T39 - TET2-mutant clonal hematopoiesis drives inflammation-associated hepatocarcinogenesis in a murine NAFLD model
Sena Alptekin, DKFZ/HI-STEM Heidelberg

09:00 - 10:30 SR1050
DZL/Breath symposium: Stem cells in lung research
Chair: *Ulrich Martin* & *Ruth Olmer*

09:00 - 09:30 T40 - Modelling type 2 alveolar epithelial cell biology and disease using patient-derived iPSC
Killian Hurley, RCSI, Ireland

09:30 - 09:45 T41 - Sotatercept as a novel therapy for pulmonary arterial hypertension: Attenuation of fibrosis and contractility in iPSC-derived smooth muscle cells
Aileen Schmidt, MH Hannover

09:45 - 10:00 T42 - The lung microvasculature promotes alveolar type 2 cell differentiation via secreted SPARCL1
Paolo Panza, UKGM, Gießen

10:00 - 10:15 T43 - Differentiation of airway epithelial basal cells into organotypic mucociliary tissues
Janna Nawroth, TU Munich

10:15 - 10:30 T44 - From injury to insight: Aberrant basaloid cells and the fortuitous discovery of a regeneration-associated cell state
Jonas Schupp, MH Hannover

09:00 - 10:30 LH23 **Pluripotency, differentiation & PSC-derived models II**
Chairs: *Micha Drukker* & *Ralf Jauch* supported by OMNI Life Science

09:00 - 09:15 T45 - Wnt/Planar cell polarity signaling orchestrates ciliogenesis and cell-cycle exit during endocrine lineage commitment
Tobias Greisle, Helmholtz Munich

09:15 - 09:30 T46 - Single-cell multiome uncovers differences in glycogen metabolism underlying species-specific speed of development
Christian Schröter, Leiden Academic Center, NL

09:30 - 09:45 T47 - NSD1 governs H3K36me2-mediated DNA methylation and drives differentiation of human iPSCs by regulating 'HIDEN' lncRNA expression
Deepika Puri, UK Aachen

09:45 - 10:00 T48 - Patient-specific gene correction in LMNA-linked cardiomyopathy using 3D hiPSC-derived heart-tissues and native myocardial slices
Eleonore Baier, TUM Munich

10:00 - 10:15 T49 - 3D transcriptomic reconstruction highlights human heart-forming organoid development as a model of early organogenesis
Liam Wilson, MH Hannover

09:00 - 10:30 HPC **Computational stem cell biology**
Chairs: *Ingmar Glauche* & *Carsten Marr*

09:00 - 09:30 T50 - Digital reprogramming decodes epigenetic barriers of cell fate changes
Eva Hörmanseder, Helmholtz Munich

09:30 - 09:45 T51 - Reinforcement learning for relapse prediction from sparse measurements in exponentially progressing disease
Arina Shelashen, TU Dresden

09:45 - 10:00 T52 - Uncovering transcriptional drivers of cell identity with generative modeling and high-throughput reprogramming
Mariia Minaeva, Helmholz Munich

10:00 - 10:15 T53 - High resolution spatio-temporal mapping reveals dedifferentiation niches in mammalian cardiac repair
Andy Shing-Fung Chan, Würzburg Institute of Systems Immunology

10:15 - 10:30 T54 - Napari-organoid-counter: An open-source tool for high-throughput organoid detection in brightfield microscopy
Christina Bukas, Helmholz Munich

10:30 - 11:00 CC & Build23 Break time / Industry exhibition supported by Qkine

11:00 - 12:30 **Concurrent strategic working group session**

11:00 - 12:30 Plenary Hall	**Technologies in stem cell research** Chairs: *Sebastian Diecke* & *Ole Pless*	supported by iotaSciences
11:05 - 11:25	S1 - Reproducible brain organoids for disease phenotyping and screening *Jakob Metzger*, Max Delbrück Center, Berlin	
11:25 - 11:45	S2 - High-content phenotypic profiling reveals subpopulation-specific modulation of human microglia by inflammasome-targeting compounds *Johannes Wilbertz*, Ksilink, Strasbourg, France	
11:45 - 12:05	S3 - Bioreactor-based innate immunocompetent hiPSC-derived neurospheroids as preclinical models for advanced therapy development *Catarina Brito*, iBet, Qeiras, Portugal	
12:05 - 12:25	S4 - BMP, MEK and WNT inhibition: A novel dSMAD-independent paradigm for rapid generation of hiPSC-derived neurons amenable to regional patterning *Peter Reinhardt*, Abbvie, Ludwighafen	
11:00 - 12:30 SR1050	**Career development** Chairs: *Marieke Essers* & *Karin Hoehne*	
	S5 - Stay calm and clear: Communication strategies for difficult situations *Nele Haasen*, konteam, Munich	
11:00 - 12:30 LH23	**Clinical trials & regulatory affairs** Chairs: *Andreas Kurtz* & *Torsten Tonn*	
11:00 - 11:30	S6 - iPSC banks for clinical use: Regulatory and ethical challenges and solutions from the perspective of EU COST Action HAPLOiPS and the Norwegian iPSC Biobank for Clinical Applications (NIBCA) *Joel C. Glover*, Norwegian Center for Stem Cell Research, Oslo, Norway	
11:30 - 12:00	S7 - tba *Joannis Mytilineos*, German National Bone Marrow Donor Registry (ZKRD), Ulm	
12:00 - 12:30	S8 - Applying haplo-selected hiPSC lines and safety switches: Significant steps towards safer ‚off-the-shelf' iPSC-based ATMPs *Gerald G. Schumann*, Paul-Ehrlich-Institut, Langen	
12:30 - 14:00 Mensa	Lunch break / Industry exhibition	supported by Stem Genomics
14:00 - 14:45 Plenary Hall	**Keynote lecture IV** K4 - From bench to bedside: Chemical reprogramming and the future of regenerative medicine Hongkui Deng, Peking University (PKU), Beijing, China Chair: *tba*	

14:45 - 15:35 **Keynote lecture V**
Plenary Hall K5 - Epigenetic mechanisms of cellular plasticity
Maria Elena Torres-Padilla, Helmholtz Munich
Chair: *Ana Martin-Villalba*

15:35 - 15:50 **Award ceremony**
Plenary Hall *Daniel Besser*
Poster awards
Travel awards
supported by Active Bioscience (AB) / PeptiGrowth (PG)

15:50 - 16:00 Closing remarks by the GSCN president
Plenary Hall *Heiko Lickert*

END OF GSCN CONFERENCE 2025

FUJIFILM
Value from Innovation

Wako



Building Partnerships in Science

FUJIFILM Wako is dedicated to advancing public health and safety through world-class reagents and support for bacterial endotoxin detection. By combining this expertise with access to JCRB's comprehensive collection of over 1,600 human and animal cell lines, we empower researchers worldwide to perform reliable research that drive scientific innovation.

Join us at GSCN 2025 to explore our cutting-edge scientific tools for cell culture and quality control applications.

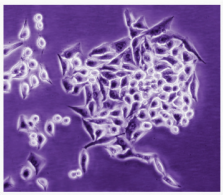
Building: 23
Stand No: 37



Quality Control Solutions

We are a comprehensive chemical supplier of high-quality excipients and fine chemicals for pharmaceutical and biopharmaceutical production in Europe, the Middle East, and Africa. Our aim is to provide customers with the highest-quality GMP raw materials, custom-tailored solutions, and sustainable quality control tests for endotoxin, non-endotoxin, and pyrogen detection.

Learn more



JCRB Cell Lines

The JCRB Cell Bank collects and distributes human and animal cultured cells worldwide, including cancer and genetically modified cell lines. All cell lines undergo testing for microbial, viral, and cross-contamination, with some characterized by karyotyping and surface markers. In partnership with global cell banks, NIBN develops culture and quality control methods to support medical and pharmaceutical research.

Learn more

FUJIFILM Wako Chemicals Europe GmbH
Fuggerstr. 12 41468 Neuss Germany
TEL: +49 (0) 2131 311-0 Fax: +49 (0) 2131 311-110
Email: info_wkeu@fujifilm.com Website: https://labchem-wako.fujifilm.com/

Speaker abstracts

13th GSCN Conference
German Stem Cell Network

15 – 17 October 2025

Helmholtz Munich
Conference Center Neuherberg

Keynote lectures: K1 – K5

K1 Advances in cell therapy for Parkinson's disease: Insights from the results of a phase 1 trial and future directions
Viviane Tabar

K2 Long-term single live cell quantification: New tools for old questions
Timm Schroeder

K3 Synthetic ex utero embryogenesis: From naive pluripotent stem cells to bona fide embryo models
Jacob Hanna

K4 From bench to bedside: Chemical reprogramming and the future of regenerative medicine
Hongkui Deng

K5 Epigenetic mechanisms of cellular plasticity
Maria-Elena Torres-Padilla

Abstract No. K1

Advances in cell therapy for Parkinson's disease: Insights from the results of a phase 1 trial and future directions

Viviane Tabar

Theresa Feng Chair in Neurosurgical Oncology Memorial Sloan Kettering Cancer Center, New York, US

The talk will provide an overview of the 2 year results of the phase 1 trial of an investigational cryopreserved, off-the-shelf dopaminergic neuron progenitor cell product (bemdaneprocel) derived from human embryonic stem (hES) cells and grafted bilaterally into the putamen of patients with Parkinson's disease. The trial enrolled 12 patients in two cohorts of high and low dose of cells injected. The trial achieved its primary objectives of safety and tolerability two years after transplantation, with no adverse events related to the cell product. At 18 months after grafting, putaminal ^{18}Fluoro-DOPA positron emission tomography uptake increased, indicating graft survival. Secondary and exploratory clinical outcomes showed improvement or stability, including improvement in the Movement Disorder Society Unified Parkinson's Disease Rating Scale (MDS-UPDRS) Part III OFF scores by an average of 23 points in the high-dose cohort. There were no graft-induced dyskinesias. A phase III randomized multi-institutional trial is ongoing.

In parallel to the clinical trials, our team is also investigating the role of the microenvironment and its impact on graft survival, using synucleinopathy models. The presentation will also provide an overview of future directions and ongoing challenges of cell therapy for Parkinson's disease.

Abstract No. K2

Long-term single live cell quantification: New tools for old questions

Timm Schroeder

ETH Zürich, Dept. of Biosystems Science and Engineering, BSSE, Basel, Switzerland

Surprisingly many long-standing questions in (stem) cell research remain disputed. One major reason is the fact that we usually analyze only populations of cells - rather than individual cells – and at very few time points of an experiment – rather than continuously.

We therefore develop imaging approaches and software to image, segment, and track cells, and to quantify e.g. divisional history, position, interaction, and protein expression or activity of all individual cells over many days and generations.

Live-cell imaging is complemented by novel large-volume multi-color 3D imaging with up to single-molecule sensitivity. Dedicated software, machine learning and computational modeling enable data acquisition, curation, and analysis. Custom-made microfluidics, optogenetics and other hardware devices improve single-cell observation, dynamic manipulation, molecular analysis, and the high-dimensional snapshot 'omics' quantification of individual cells with known history, kinship and dynamics.

The resulting continuous single-cell data is used for analyzing the dynamics, interplay, and functions of signaling pathway and transcription factor networks in controlling the fate decisions of self-renewing and differentiating hematopoietic, pluripotent, neural stem and intestinal stem and progenitor cells and normal and synthetic embryos.

Abstract No. K3

Synthetic ex utero embryogenesis: From naive pluripotent stem cells to bona fide embryo models

Jacob Hanna

Weizmann Institute of Science, Rehovot, Israel

The identity of somatic and pluripotent cells can be epigenetically reprogrammed and forced to adapt a new functional cell state by different methods and distinct combinations of exogenous factors. The aspiration to utilize such in vitro reprogrammed pluripotent and somatic cells for therapeutic purposes necessitates understanding of the molecular mechanisms of reprogramming and differentiation and elucidating the extent of equivalence of the in vitro derived cells to their in vivo counterparts. In addition to the latter, in my presentation, I will present my group's recent advances toward understanding these fundamental questions and further detail our ongoing efforts to generate developmentally unrestricted human & mouse naive pluripotent cells with embryonic and extra-embryonic developmental potential. I will expand on new avenues for utilizing custom made electronically controlled ex utero platforms and optimized conditions for growing natural mammalian embryos ex utero for extended periods capturing development from pre-gastrulation until advanced organogenesis, for better studying of stem cell transitions during embryogenesis and organogenesis. I will detail how the latter platforms offered an exclusive technical platform to demonstrate and unleash the self-organizing capacity of mouse naïve PSCs to generate post-gastrulation synthetic bona fide Structured, Synthetic & Stem-cell-based whole Embryo Models (SEMs) with both embryonic and extra-embryonic compartment ex utero, as well as our ability to extend these findings with naïve human PSCs and generate complete structured day 14 human developmental models and beyond. Collectively, I will be highlighting prospects for revolutionary platforms for advancing human disease and embryogenesis ab initio developmental modelling.

Abstract No. K4

From bench to bedside: Chemical reprogramming and the future of regenerative medicine

Hongkui Deng

Peking University, China & Peking-Tsinghua Center for Life Sciences, China

The use of cell intrinsic components, including oocyte cytoplasm and transcription factors, can enforce somatic cell reprogramming to pluripotent stem cells. By contrast, chemical stimulation by exposure to small molecules offers a new approach that can manipulate cell fate in a simple and highly controllable manner. In 2022, we demonstrated that small molecules can effectively reprogram human somatic cells to pluripotent stem cells. Our chemical approach provides a new platform for the generation and application of human pluripotent stem cells in biomedicine.

One such example is the generation of stem cell-derived pancreatic islets for cell replacement therapy for diabetes. We established a robust protocol that generates functional pancreatic islet cells from hCiPSCs. When transplanted, these hCiPSC-derived islets effectively ameliorated diabetes and restored robust endogenous insulin secretion in diabetic nonhuman primates, demonstrating their therapeutic potential against diabetes. Recently, chemically-induced stem cell-derived islets (CiPSC-islets) were transplanted underneath the abdominal anterior rectus sheath of a patient, resulting in rapid reversal of diabetes. The patient achieved all pre-specified primary and secondary clinical study endpoints at 2 years follow-up. The results from the first patient suggest feasibility of this treatment modality and warrant its further exploration.

Abstract No. K5

Epigenetic mechanisms of cellular plasticity

Maria-Elena Torres-Padilla

Institute of Epigenetics & Stem Cells, Helmholtz Munich

Cells at the earliest stages of development display the highest level of plasticity throughout the mammalian life cycle. In mammals, the terminally differentiated sperm and oocyte fuse to create a totipotent zygote upon fertilisation. The mechanisms underlying the epigenetic reprogramming towards totipotency are not fully understood and the molecular features of totipotent cells remain scarce. Embryonic cells of the early embryo remain totipotent only for a restricted time window. During this time, embryonic cells are characterised by an atypical chromatin structure, a high degree of chromatin plasticity and reactivation of specific families of retrotransposons. We have studied several layers of epigenetic regulation and chromatin structure in early mouse embryos. Accordingly, we have begun to decipher key molecular regulators of chromatin plasticity and epigenome establishment, including the potential regulatory role of transposable elements in the embryo and how heterochromatin first forms during this reprogramming process. We will present our latest results indicating a role for chromatin integrity in promoting epigenetic reprogramming and sustaining molecular features of totipotent cells in vivo. Finally, we will discuss the potential conservation of these findings across mammals beyond the mouse model.

Presidential Symposium lectures: PS1 – PS4

GSCN Presidential Invite

PS1 Translation of tissue engineered heart repair
Wolfram-Hubertus Zimmermann

GSCN 2025 Young Investigator Award

PS2 Molecular regulation of mammalian dormancy
Aydan Bulut-Karslioglu

GSCN 2025 Hilde Mangold Award

PS3 Harnessing RNA modifications for cancer therapies
Michaela Frye

GSCN Publication of the Year 2024 - 2025 Award

PS4 The role of safeguard repressors in cancer
Bryce Lim

Abstract No. PS1: GSCN Presidential Invite

Translation of tissue engineered heart repair

Wolfram-Hubertus Zimmermann

Institute of Pharmacology and Toxicology, University Medical Center Göttingen & German Center for Cardiovascular Research (DZHK), partner site Lower Saxony

Engineered Heart Muscle (EHM) with properties of juvenile myocardium can be constructed from pluripotent stem cell-derived cardiomyocytes and stromal cells. To facilitate EHM self-organization, cells are embedded in a collagen hydrogel and subjected to mechanical conditioning. Preclinical testing in a homologous Rhesus macaque model confirmed that induced pluripotent stem cell-derived EHM allograft implantation is feasible, safe, and efficacious under an optimized immune suppression protocol. The knowledge gained from late preclinical testing in Rhesus macaques informed the design of the first-in-patient “Safety and Efficacy of Induced Pluripotent Stem Cell-derived Engineered Human Myocardium as Biological Ventricular Assist Tissue in Terminal Heart Failure (BioVAT-HF)” Phase I/II clinical trial (NCT04396899). First proof of remuscularization in a patient subjected to a successful heart transplantation 3 months after EHM implantation was obtained, demonstrating the feasibility of remuscularization in humans. Experience from late preclinical and early clinical studies will be discussed.

Abstract No. PS2: GSCN 2025 Young Investigator Award

Molecular regulation of mammalian dormancy

Aydan Bulut-Karslioglu

Max Planck Institute for Molecular Genetics, Berlin

Dormancy is a conserved mechanism that preserves cellular potential in development and tissue homeostasis. Mammalian embryos can enter diapause, a reversible dormant state, but the molecular basis of this adaptation has remained unclear. Our recent studies reveal that dormancy is not a passive process but requires extensive metabolic, transcriptional, and epigenetic rewiring.

At the metabolic level, we found that dormant embryos switch their energy use to fatty acid oxidation under the control of FOXO1. L-carnitine supplementation enforces this switch, extends dormancy, and points to lipid metabolism as a shared regulator across tissues. At the chromatin level, we showed that TET enzymes maintain pluripotency by protecting regulatory elements from silencing. Loss of TET activity destabilizes pluripotency during dormancy, whereas enhanced activity improves embryo survival. Finally, we identified a requirement for microRNAs in the dormancy transition. Although dispensable during steady-state preimplantation development, combinatorial miRNA activity is essential for diapause entry and survival, linking nutrient sensing to the maintenance of pluripotent cells.

Together, our findings position metabolism, epigenetic safeguarding, and transcriptional regulation as layers of a coordinated dormancy program. In my talk, I will discuss these findings in greater detail and elaborate on their broader implications.

Abstract No. PS3: GSCN 2025 Hilde Mangold Award

Harnessing RNA modifications for cancer therapies

Michaela Frye

DKFZ, Heidelberg

The frequent dysregulation of N6-methyladenosine (m6A), the most abundant internal mRNA modification, creates exploitable vulnerabilities in cancer, as its function is highly context-dependent. Progress is, however, limited by a fragmented and sometimes contradictory understanding of m6A biology. To address this, I will define universal versus cancer-specific m6A functions in head and neck squamous cell carcinoma (HNSC), an aggressive cancer in which m6A biology is poorly understood. I will present mechanistic examples showing how m6A shapes gene transcription and how its depletion impairs DNA damage and repair responses, leading to the activation of innate immunity pathways. These m6A-dependent mechanisms also intersect with the cancer cell's energy metabolism, enhancing metabolic plasticity. Finally, I will integrate these insights to explore how components of the m6A machinery can be leveraged as a part of novel targeted therapies in HNSC.

Abstract No. PS4: GSCN Publication of the Year 2024 - 2025 Award

The role of safeguard repressors in cancer

Bryce Lim[1,*], *Aryan Kamal*[2], *Borja Gomez Ramos*[1], *Juan Adrian Segarra*[1], *Ignacio Ibarra*[2], *Lennart Dignas*[1], *Tim Kindinger*[1], *Kai Volz*[1], *Mohammad Rahbari*[1], *Nuh Rahbari*[1], *Eric Poisel*[1], *Kanela Kafetzopoulou*[1], *Lio Böse*[1], *Marco Breinig*[1], *Danijela Heide*[1], *Suchira Gallage*[1], *Jose Barragan Avila*[1], *Hendrik Wiethoff*[1], *Ivan Berest*[2], *Sarah Schnabellehner*[3], *Martin Schneider*[1], *Jonas Becker*[4], *Dominic Helm*[1], *Dirk Grimm*[1], *Taija Mäkinen*[3], *Darjus Tschaharganeh*[1], *Mathias Heikenwalder*[1], *Judith Zaugg*[2], and *Moritz Mall*[1]

[1] DKFZ, Heidelberg
[2] EMBL, Heidelberg
[3] Uppsala University, Sweden
[4] Heidelberg University

*Presenting author

Cell fate plasticity, a developmental phenomenon, emerges as a critical hallmark of cancer when dysregulated. While loss of master regulators such as Pax5 that activate lineage-specific genes can cause cancer, the role of lineage-specific repressors in preventing unwanted plasticity in cancer remains poorly understood. We previously found that the neuron-specific repressor MYT1L could suppress many non-neuronal gene programs to safeguard neuronal cell fate. Our work introduces the concept of “safeguard repressors,” lifelong-expressed cell type-specific transcriptional repressors that maintain cell identity to block cancer. We developed novel computational tools alongside single-cell datasets to identify and validate these repressors in liver and brain cancers, both known for their high plasticity.

Here, we predict safeguard repressor candidates for 18 cell types that can suppress many alternative fates to safeguard the desired cell identity. We validated hepatocyte-specific candidates using cell fate reprogramming, revealing that Prospero homeobox protein 1 (PROX1) enhanced hepatocyte identity and suppressed several alternate cell identities. Mechanistically, direct repression of PROX1 target genes, including alternate fate master regulators Prrx1 and Pparg, promoted hepatocyte fate. Deleting Prox1 impaired hepatocyte formation and caused the induction of alternate fate markers. In liver cancer, PROX1 was found to preserve hepatocyte identity by repressing alternative fate regulators. Overexpression of PROX1 in hepatocellular carcinoma mouse models blocked tumor initiation, progression, and notably extended survival. Conversely, PROX1 depletion caused hepatocyte fate loss and promoted transdifferentiation to cholangiocarcinoma. Clinically, low PROX1 expression correlated with poor prognosis in liver cancer patients, underscoring its protective role against cell fate plasticity and oncogenic transformation.

Our findings highlight that dedicated safeguard repressors like PROX1 and MYT1L are vital for suppressing cellular plasticity that fuels cancer. Further studies, supported by advanced computational strategies as developed here, could identify similar mechanisms across other cancers, presenting new avenues to combat cancer progression by stabilizing cell identity.

Selected presentations

13th GSCN Conference
German Stem Cell Network
15 – 17 October 2025

Selected presentations: T01 – T54

T01 Manufacturing and treatment with CAR effector cells: From personalized to iPS-based off the shelf products and from viral to non-viral gene transfer
Ulrike Koehl

T02 Maximizing gene editing efficiency for enhanced clinical grade iPSCs via sequential factor delivery
Thomas Berger

T03 Off the shelf immune-shielded stem cell-derived macrophages for clinical applications
Stefanie Pfaender

T04 Prototypical ten-week end-to-end process for autologous Parkinson's disease cell therapy
Diana Haberhausen

T05 Enhancing stem cell derived beta cell survival in hypoxia and nutrient deprivation with small molecules
Väinö Lithovius

T06 Mapping the transcriptome and DNA-methylome at the single-cell level decodes stemness across the young and injured brain
Ana Martin-Villalba

T07 Manipulation of global protein translation rates and stem cell self-renewal by CRISPR mediated activation of rRNA transcription
Stefan Stricker

T08 Slit–Robo signaling controls early liver zonation during fetal development
Dominic Grün

T09 Unveiling human cardiac regenerative mechanisms in iPSC-derived epicardioids
Tobias Ellinger

T10 Human-specific morphoregulatory signatures in basal radial glia characterize neocortex evolution
Mareike Albert

T11 Epicardioids: Bridging cardiac development with disease and therapy
Alessandra Moretti

T12 AI-guided protein design of synthetic growth factor mimetics
Thomas Schlichthärle

T13 Enhanced fibrosis modeling in human skin organoids through hiPSC-derived macrophage integration
Amelie Reigl

T14 Heart organoids and engineered heart tissue as hiPSC-derived in vitro models for Fabry cardiomyopathy
Anna Reinelt

T15 iPSC-driven drug discovery identifies sildenafil as a drug candidate for the mitochondrial disease Leigh syndrome
Annika Zink

T16 Blood stem cell expansion: Technology to biology
Adam Wilkinson

T17 C57BL/6 Kit-mutant immunodeficient mouse models uncover mechanism of successful hematopoietic stem cell xenotransplantation
Julia Fröbel

T18 From codons to blood: A new perspective on hematopoietic differentiation
Marius Külp

T19 Multimodal omics identify an age resilient multipotent progenitor population that preserves youthful identity and function in the hematopoietic system
Mari Carmen Romero-Mulero

T20 Development of accessible ex vivo culture models of the human hematopoietic bone marrow niche
Sigrid Uxa

T21 Deep immune-phenotyping of HLA-homozygous iPS-cardiomyocytes
Dirk Strunk

T22 Biological-engineering and application of a myocardial patch
Joseph Petrus Gerardus Sluijter

T23 Large-scale production of human pluripotent stem cell–derived cardiomyocytes
Kevin Cyrys

T24 Automated myocardial cell injection
Raphael Mönkemöller

T25 Developing a safe and immune-evasive stem cell source for off-the-shelf therapeutic products
Andras Nagy

T26 Lineage single-cell multi-omics identify novel regulators and barriers in iPSC reprogramming
Angeliki Spathopoulou

T27 You shall not pass - functional pluripotency testing using hiPSCore
Jochen Dobner

T28 PluripotencyScreen – quantification of early cell fate decisions of iPSC based on DNA methylation
Kira Zeevaert

T29 Does gene dosage influence cell fate during human gastrulation?
Theresa Gross-Thebing

T30 Modulating reactive astrocytes in Alzheimer's disease*
Bastian Zimmer

T31 Evaluating the potential of Bruton's tyrosine kinase inhibitors to modulate microglial inflammation to attenuate multiple sclerosis progression
Alica Blenkle

T32 A reproducible human brain tissue model to study physiological and disease-associated microglia phenotypes
Dominik Paquet

T33 Integration of CNTNAP2-deficient neurons into defined E/I networks unmasks functional pathophenotypes
Jasmin Aicher

T34 Capturing disease severity in LIS1-lissencephaly reveals proteostasis dysregulation in patient-derived forebrain organoids
Matteo Gasparotto

T35 Tracing clonal architecture and differentiation trajectories in cancer using single cell barcoding
Michael Rieger

T36 Role of aging and environmental stress in the decline of lung stem cell function and regenerative capacity
Maja Funk

T37 Investigating aging and telomere biology in lung disease using induced pluripotent stem cell derived models
Mari Ozaki

T38 High fat diet affects the stromal niche signaling in the porcine intestine
Falk Janos Farkas

T39 TET2-mutant clonal hematopoiesis drives inflammation-associated hepatocarcinogenesis in a murine NAFLD model
Sena Alptekin

T40 Modeling type 2 alveolar epithelial cell biology and disease using patient-derived iPSC
Killian Hurley

T41 Sotatercept as a novel therapy for pulmonary arterial hypertension: attenuation of fibrosis and contractility in iPSC-derived smooth muscle cells
Aileen Schmidt

T42 The lung microvasculature promotes alveolar type 2 cell differentiation via secreted SPARCL1
Paolo Panza

T43 Differentiation of airway epithelial basal cells into organotypic mucociliary tissues
Janna Nawroth

T44 Aberrant basaloid cells and the fortuitous discovery of a regeneration-associated cell state
Jonas Schupp

T45 Wnt/Planar cell polarity signaling orchestrates ciliogenesis and cell-cycle exit during endocrine lineage commitment
Tobias Greisle

T46 Single cell multiome uncovers differences in glycogen metabolism underlying species-specific speed of development
Christian Schröter

T47 NSD1 governs H3K36me2-mediated DNA methylation and drives differentiation of human iPSCs by regulating 'HIDEN' lncRNA expression
Deepika Puri

T48 Patient-specific gene correction in LMNA-linked cardiomyopathy using 3D hiPSC-derived heart-tissues and native myocardial slices
Eleonore Baier

T49 3D transcriptomic reconstruction highlights human heart-forming organoid development as a model of early organogenesis
Liam Wilson

T50 Digital reprogramming decodes epigenetic barriers of cell fate changes
Eva Hörmanseder

T51 Reinforcement learning for relapse prediction from sparse measurements in exponentially progressing disease
Arina Shelashen

T52 Uncovering transcriptional drivers of cell identity with generative modeling and high-throughput reprogramming
Mariia Minaeva

T53 High resolution spatio-temporal mapping reveals dedifferentiation niches in mammalian cardiac repair
Andy Shing-Fung Chan

T54 Napari-organoid-counter: An open-source tool for high-throughput organoid detection in brightfield microscopy
Christina Bukas

Abstract No. T01

Manufacturing and treatment with CAR effector cells: From personalized to iPS-based off the shelf products and from viral to non-viral gene transfer

Ulrike Koehl

Fraunhofer Institute for Cell Therapy and Immunology (IZI) & Institute of Clinical Immunology, University Hospital and University of Leipzig

Chimeric antigen receptor (CAR) T cell therapy has transformed the treatment landscape for relapsed/refractory hematological diseases, providing also a promising option for patients with solid tumours and autoimmune diseases. Starting with a short overview about ongoing CAR cell trials, next the reproducible manufacture of high-quality clinical-grade CAR T cells will be discussed. With increasing numbers of patients, this is becoming an ever-greater challenge. New processing techniques, quality-control mechanisms, and logistic developments are required to meet both medical needs and regulatory restrictions. Still, personalized manufacturing is time consuming and expensive. Results with automated manufacturing gives rise for improvement in both centralized and decentralized manufacturing units. However, a modular, open and transferable system with AI-mediated robotics and digital control as well as the respective automated documentation of all in process parameters is still missing in order to address 100-fold more patients. In addition, there are no harmonized rules for patient selection regarding the leukapheresis starting material and surrogate marker are nearly missing to predict production failure. In several cases, failure in manufacturing appears because the patients are heavily pre-treated and this can influence the fitness of the cells substantially. Allogeneic CAR effector cells, such as mature or iPS-derived CAR NK cells play an increasing role to overcome the limitations in autologous CAR T cell manufacturing. Next, the switch from lentiviral to AAV or non viral gene transfer, based on sleeping beauty and mRNA/LNP technologies has opened a platform for improved safety and decreased manufacturing costs as well as for direct in vivo therapy. In summary both, improvement in manufacturing as well as concepts to use allogeneic effector cells with non-viral gene transfer are necessary to overcome current limitations.

Abstract No. T02

Maximizing gene editing efficiency for enhanced clinical grade iPSCs via sequential factor delivery

*Thomas Berger**, *Elitsa Borisova*, and *Boris Greber*

Catalent Düsseldorf GmbH

*Presenting author

Human induced pluripotent stem cells (iPSCs) are a promising platform for cell therapy, but immune rejection due to HLA variability remains a major challenge. Autologous iPSC therapies avoid this issue but face high costs and extended production timelines. HLA-homozygous lines improve compatibility for off-the-shelf use, yet a broad population coverage would require a large and diverse panel. A more scalable strategy is to generate universal allogeneic iPSCs via targeted gene editing to reduce immunogenicity. Despite putative safety concerns, this approach can be strengthened by integrating fail-safe systems and immunomodulatory transgenes to generate universal donor cells.

Using Catalent's GMP-compatible iPSC lines, we systematically optimized CRISPR-Cas9 and Cas12a ribonucleoprotein (RNP)-based genome editing protocols. Based on sequential delivery of donor plasmids and RNPs as pivotal element, this approach enables targeted knock-ins of full-length transgenes with efficiencies exceeding 30%, all without the need for cell sorting, antibiotic selection, or viral vectors – enabling the identification of positive clones by random picking at small scale. To demonstrate utility of the platform, we have generated and characterized iPSC lines with disruption of HLA class I (and II), along with knock-ins of an inducible caspase-9 (iCasp9) suicide gene and/or CD47 overexpression. The resulting edited lines maintained genomic stability, pluripotency, and differentiation potential throughout. These edits confer immune evasion properties and allow for pharmacologically controlled elimination of transplanted cells in the event of adverse reactions. This integrated platform provides a versatile and scalable system for producing gene-edited, GMP-grade iPSCs with enhanced safety and functionality, supporting the advancement of allogeneic iPSC-based therapies.

Abstract No. T03

Off the shelf immune-shielded stem cell-derived macrophages for clinical applications

*Stefanie Pfaender**, *Kathrin Haake*, *Andrej Schebet*, *Philine Scheinpflug*, *Alica Hinkelmann*, *Elisabeth Derksen*, *Farah Aqel*, *Jonas Klaas*, *Tim Magiera*, *Sebastian Kirsch*, *Anais Duval*, *Philip Hublitz*, *Michela Mirenda*, *Martin Briscadieu*, *Michael Esquerre*, *Michael Epstein*, *Katrin Ridders*, *Ariane Walsh*, *Andreas Scheel*, and *Matthias Austen*

Evotec SE, Göttingen

*Presenting author

Macrophages have emerged as promising candidates for cell-based immunotherapies against solid malignancies and fibrotic disease due to their high tissue-infiltrating capacity, intrinsic plasticity and central role in the immune response. Macrophages can be enhanced further for clinical applications via genetic engineering. This includes the introduction of chimeric antigen receptors (CARs) for antigen-specific targeting, the deletion of checkpoint inhibitory proteins to prevent inhibition in the target tissue or the deletion/introduction of HLA-molecules to enhance persistence by cloaking cells from the host immune system. Contrary to donor-derived macrophage therapies, where availability and quality of cells and amenability to genetic engineering pose major challenges, macrophages differentiated from clonal genetically engineered iPSC are readily available with a defined genotype, phenotype and purity.

We generated GMP-iPSC line-derived genetically engineered lines by deleting the immune checkpoint inhibitor SIRPα, by introducing a CAR and immune-shielding modifications using a targeted approach. We further developed a scalable 3D xeno-free in vitro differentiation process allowing large-scale production of genetically engineered immune-shielded functional CAR-macrophages for clinical applications.

Engineered iPSC were enriched and isolated clones were quality-controlled for correct transgene insertion, genetic integrity and pluripotency prior to banking. iPSC clones were differentiated feeder- and serum-free in stirred tank bioreactors into functional macrophages with a purity of >90% and a ~10fold expansion/iPSC input.

SIRPα-deletion led to increased phagocytic activity and killing capacity in differentiated macrophages. Similarly, PoC was established for CAR expression and function associated with enhanced phagocytosis and killing of relevant target cells.

Taken together, we have engineered clonal iPSC lines with CARs, SIRPα-deletion and immune evasion edits for customized, highly pure, off-the-shelf CAR-macrophage therapies against solid tumors or lung/liver fibrosis, differentiated in a xeno-free, GMP-compatible and scalable in vitro process.

Abstract No. T04

Prototypical ten-week end-to-end process for autologous Parkinson's disease cell therapy

Diana Haberhausen[1,2,*], *Hannah Johannsen*[1], *Jana Bonsberger*[3], *Andreas Bosio*[1], *Ullrich Wüllner*[2], *Malin Parmar*[3], and *Sebastian Knöbel*[1]

[1] Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
[2] UKB Universitätsklinikum Bonn
[3] Lund University, Sweden

*Presenting author

Parkinson's Disease (PD) is the second-most prevalent neurodegenerative disorder characterized by progressive degeneration of dopaminergic neurons, leading to motor dysfunction and cognitive decline. Due to demographic change and increasing numbers of patients, PD gains importance and novel treatment options for PD are urgently needed. Cumulative research has shown that stem cell-based therapies offer considerable promise but still fundamental challenges remain unresolved. With focus on autologous cell therapy, functional and phenotypic comparability between cell products as well as batch-to-batch consistency is mandatory to maximize the use of iPSC-derived mesencephalic dopaminergic (mDA) progenitors in the clinic. Based on skin biopsies of healthy donors, we established a rapid and robust iPSC manufacturing process that generates iPSC stocks within seven weeks and includes comprehensive quality control to select bona-fide iPSCs for downstream differentiation. We optimized existing mDA differentiation protocols, so that after 16 days of differentiation, iPSC-derived mDA progenitors of several iPSC lines exhibited the desired phenotype. We could provide proof of concept that mDA cells from two different donors were functional and normalized amphetamine-induced rotation in a 6-OHDA rat model. For ex vivo phenotyping of preclinical animal models we developed imaging solutions to describe first, the spatial projection and quantification of transplanted cells using 3D light-sheet imaging and second, the graft composition with cyclic immunofluorescent imaging in 2D. To facilitate scalability and standardization, we successfully transferred key elements of the iPSC generation and mDA differentiation workflow to semi-automated platforms, paving the way towards future clinical translation. Moreover, we transferred our workflow to relevant patient-derived samples. Thus, we generated PD patient-derived iPSCs, performed stringent quality control and differentiated bona-fide iPSC lines into mDA progenitors. Our data provide evidence that by keeping factors like somatic cell type, reprogramming method, and culture platform constant, standardization and comparability of the cells is highly favoured, minimizing batch-to-batch variability.

Abstract No. T05

Enhancing stem cell derived beta cell survival in hypoxia and nutrient deprivation with small molecules

Väinö Lithovius[1,*], *Nicole Rogers*[1], *Christos Karampelias*[1], and *Heiko Lickert*[2]

[1] Helmholtz Diabetes Center, Helmholtz Munich
[2] Institute of Diabetes and Regeneration, Helmholtz Munich

*Presenting author

Type 1 diabetes is caused by autoimmune destruction of the insulin secreting beta cells of the pancreatic islets. It is typically managed with exogenous insulin replacement, but rarely by replacing the lost beta cells with islet transplantation. This reconstitutes endogenous insulin secretion and is associated with improved treatment outcomes. Availability of islet transplantation is restricted by low availability of donor islets, which is further exacerbated by large losses of the islets upon engraftment, ranging from 60 to 90%, before they connect to recipient vasculature. This necessitates multiple donors per recipient. Currently, islets can be differentiated *in vitro* from stem cells (SC-islets) improving islet availability and are in clinical trials. However, also SC-islets are susceptible to hypoxia and loss of nutrient supply. Beta cell loss in SC-islets can lead to high treatment cost and outcome variability with a risk of enriching for unwanted off-target cells arising as byproducts of differentiation.

We aimed to find interventions that could mitigate the loss of beta cells upon engraftment. For this, we established in vitro conditions mimicking the immediate post-transplantation period by reducing oxygen and nutrient availability. In these conditions, we conducted a screen of 190 molecules approved for human use using SC-islets modified to express either mCherry or nanoluciferase in the insulin locus, enabling high-throughput testing of beta cell function and survival. We identified compound classes that consistently enhanced beta cell survival. By the time of the conference, we will have conducted detailed hit-confirmation assays of the top hits and their combinations paving the way for *in vivo* validation studies.

In conclusion, we have established conditions that enable high-throughput screening of small molecules and identified small molecules protecting beta cells in hypoxia and nutrient deprivation *in vitro*.

Abstract No. T06

Mapping the transcriptome and DNA-methylome at the single-cell level decodes stemness across the young and injured brain

Ana Martin-Villalba

German Cancer Research Center (DKFZ), Heidelberg

Life choices are not just about what we want to do but as well about who we want to be. Similarly, performance and fate underlie a decision-making process in the life of a stem cell within the adult brain. My lab focuses on understanding the mechanisms underlying stem cell choices in the healthy, injured and aging brain. In order to study stem cells globally within the CNS in a living organism, we have taken a multiomics approach that combines profiling of the transcriptomes, chromatin accessibility and methylome of single cells along the neural stem cell's journey into a differentiated state in the healthy and ischemic brain. In my talk I will discuss what we have learned about neural stem cell heterogeneity, how methylome controls fate and about the interplay of these three molecular layers along the different maturation stages.

Abstract No. T07

Manipulation of global protein translation rates and stem cell self-renewal by CRISPR mediated activation of rRNA transcription

Stefan Stricker

LMU Munich

Rates of protein translation change during development and differ remarkably across cell types. Consequently, cellular translation rates are suspected to play a role in a number of integral cellular processes. Expression levels of ribosomal RNAs are of particular interest, not only because those have an essential role in protein translation, but also because their mis-regulation has been implicated in a number of diseases, such as cancer. However, direct testing of the functional relevance of rRNA levels have proven to be challenging, as options to increase rRNA levels and/or translational rates are limited due to the high abundance of rRNAs and the large number of rDNA genes. Here we report the development and use of TAPIR (Targeted Activation of Protein Translation), which is based on CRISPR transactivators and gRNAs designed to increase cellular levels of ribosomal RNAs. Through this manipulation, 47S rDNA clusters become transcriptionally induced, but rRNA processing remains intact, resulting in highly elevated levels of 18S and 28S rRNAs. Induction of rRNA results in extended nucleoli and in heightened protein translation rates, even in already fast dividing cells. Finally, we show for neural stem cells, that increasing translation rates stimulate stem cell self-renewal in vitro. Moreover, by transferring TAPIR in vivo similar effects are apparent, as stem cells of the developing forebrain are dividing more frequently and alter fate decisions, when rRNA levels are elevated. By this we can demonstrate for the first time that rRNA expression levels regulate protein translation levels directly and protein translation rates are key regulators of mammalian stem cell behavior.

Abstract No. T08

Slit–Robo signaling controls early liver zonation during fetal development

Gosia Golda[1,2], *Lisa Marschhäuser*[1], *Judith Schaf*[1], *Nadine Vornberger*[1], and *Dominic Grün*[1,*]

[1] Institute of Systems Immunology, University of Würzburg
[2] Max Planck Institute of Immunobiology and Epigenetics, Freiburg

*Presenting author

A complex interplay of intrinsic transcriptional programs and extrinsic signaling cues governs the spatial organization, lineage specification, and maturation of hepatoblasts during liver development. A key feature of liver physiology is metabolic zonation, in which hepatocytes acquire distinct identities along the porto-central axis. While adult liver zonation is shaped by postnatal gradients of Wnt/β-catenin signalling, oxygen, and nutrients, the developmental mechanisms that spatially position hepatoblasts to interpret these cues remain poorly understood.

We analyzed ~50,000 single-cell transcriptomes from mouse embryonic livers (E8.5–E15.5) and identified migrating hepatoblasts with early transcriptional biases toward pericentral or periportal identity. These subpopulations differentially express Slit–Robo guidance receptors, with Robo1 associated with pericentral-biased and Robo2 with periportal-biased gene expression, suggesting a developmental guidance role in hepatic patterning. Using organoid cultures, we confirmed functional differences between these populations, reflected in distinct organoid-forming capacities.

These differences parallel spatial signaling environments in the embryonic liver. Slit2, a Robo ligand, is expressed in periportal endothelial cells, supporting a model in which Slit–Robo signaling functions as a guidance system that pre-patterns hepatoblasts along the porto-central axis. Conditional deletion of Robo1 and Robo2 in hepatoblasts disrupts this pattern, resulting in aberrant zonation and liver dysfunction in adult mice, highlighting that early spatial positioning is critical for establishing proper metabolic architecture.

We also identified a transcriptionally similar hepatoblast population in human fetal liver, with conserved expression of zonation markers and Slit–Robo genes, indicating an evolutionarily conserved early patterning mechanism.

Together, our findings uncover a previously unrecognized role for Slit–Robo signaling in hepatoblast positioning and zonation. They challenge the view of zonation as purely a postnatal process and emphasize the importance of developmental guidance cues in shaping liver architecture, with implications for understanding liver disease and regeneration.

Abstract No. T09

Unveiling human cardiac regenerative mechanisms in iPSC-derived epicardioids

Tobias Ellinger[1,*], *Luis Felipe Monge Mora*[1], *Niklas Petzold*[2], *Anne Dueck*[2], *Ulrich Martin*[3], *Tatjana Dorn*[1], *Karl-Ludwig Laugwitz*[1], and *Alessandra Moretti*[1]

[1] First Department of Medicine, Cardiology, TUM Klinikum Rechts der Isar, Munich
[2] Institute of Pharmacology and Toxicology, Technical University Munich (TUM)
[3] Department of Cardiothoracic, Transplantation and Vascular Surgery (HTTG), Hannover Medical School

*Presenting author

Differently from the embryonic heart, the adult human heart exhibits a limited capacity for regeneration following injury, posing a significant clinical challenge. To investigate human-specific regenerative processes, we utilized human induced pluripotent stem cell (hiPSC)-derived cardiac organoids, specifically "epicardioids," which show self-organization and patterning of functional epicardium and ventricular myocardium resembling the embryonic ventricular wall (Meier et al., 2023).

Employing a genetic cell ablation system within epicardioids, we observed a robust epicardial activation response. However, this alone was insufficient to support true myocardial regeneration. Strikingly, co-culture with macrophages enhanced regenerative features, including increased endothelial cell formation, extracellular matrix remodeling, and cardiomyocyte mobilization toward the injury site.

Our findings highlight the pivotal role of macrophages in modulating regenerative responses in human cardiac tissue models and suggest new strategies to unlock the heart's latent regenerative capacity. This work demonstrates the value of organoid systems for studying complex repair mechanisms in a human-specific context.

Meier, A.B., Zawada, D., De Angelis, M.T. et al. Epicardioid single-cell genomics uncovers principles of human epicardium biology in heart development and disease. Nat Biotechnol 41, 1787–1800 (2023). https://doi.org/10.1038/s41587-023-01718-7

Abstract No. T10

Human-specific morphoregulatory signatures in basal radial glia characterize neocortex evolution

Theresa M. Schütze[1], *Nora Ditzer*[1], *Silvia Vangelisti*[2], *Annika Kolodziejczyk*[1], *Emanuele Capra*[3], *Ilaria Chiaradia*[4], *Jula Peters*[5], *Maximilian Krause*[1], *Christina Eugster*[5], *Razvan P. Derihaci*[1], *Cahit Birdir*[1], *Ulrich Martin*[6], *Pauline Wimberger*[1], *Katherine R. Long*[7], *Madeline Lancaster*[4], *Nereo Kalebic*[3], *Boyan Bonev*[2], and *Mareike Albert*[1,*]

[1] TUD Dresden University of Technology
[2] Helmholtz Munich
[3] Human Technopole, Milan, Italy
[4] MRC Laboratory of Molecular Biology, Cambridge, UK
[5] Max Planck Institute of Molecular Cell Biology and Genetics (MPI-CBG), Dresden
[6] Hannover Medical School
[7] King's College London, UK

*Presenting author

As the seat of our cognition, the human neocortex is an object of immense fascination. Human neocortex expansion during evolution has been attributed to an increase in the proliferative capacity of neural progenitor cells during development, particularly basal radial glia (bRG). Despite their evolutionary relevance, the genomic changes driving human-specific bRG biology remain uncharacterized. We used comparative chromatin and transcriptional profiling of neural progenitor cells isolated from gorilla, chimpanzee and human cerebral organoids to identify cis-regulatory elements that have gained activity in humans. Focusing specifically on bRG, we discovered that morphoregulatory enhancer activity and gene expression signatures distinguish human bRG from other great apes. Functional analysis of the morphoregulatory genes FAM107A and CNGA3 in human organoids revealed that these genes are required for the morphological complexity of human bRG. Taken together, our interspecies comparison of basal radial glia suggests that human-specific morphoregulatory signatures characterize neocortex evolution.

Abstract No. T11

Epicardioids: Bridging cardiac development with disease and therapy

Alessandra Moretti

Department of Medicine I: Cardiology, TUM Klinikum Rechts der Isar, Technical University of Munich & DZHK (German Center of Cardiovascular Research), Munich Heart Alliance

Human pluripotent stem cell–derived organoids offer powerful opportunities to model cardiac development and disease. We recently established *epicardioids*—self-organizing 3D tissues that recapitulate key aspects of the human ventricular wall, including a myocardial core surrounded by an epicardial-like layer. This architecture makes epicardioids uniquely suited for studying interactions between myocardium and epicardium during heart development, as well as for modeling pathological processes such as fibrosis, remodeling, and cardiac hypertrophy. This talk will highlight how epicardioids can serve as a versatile platform for disease modeling and preclinical drug discovery, providing new avenues to bridge stem cell biology with translational cardiac research.

Abstract No. T12

AI-guided protein design of synthetic growth factor mimetics

Thomas Schlichthärle[1,*], *Natasha Edman*[2], *Aerin Yang*[3], *Ashish Phal*[2], *Damien Detraux*[2], *Rachel Redler*[4], *David Johnson*[5], *Hannele Ruohola-Baker*[2], *Christopher Garcia*[3], and *David Baker*[2]

[1] TU Munich
[2] University of Washington, Seattle, US
[3] Stanford University, US
[4] New York University, US
[6] Seattle Children's Hospital, US

*Presenting author

Recent advances in AI-guided protein design now enable the computational generation of novel binding domains for receptor targets with high experimental success rates. These domains can be precisely engineered to cluster specific receptor subtypes, resulting in efficient pathway activation and fine-tuned modulation of downstream signaling. In a first study, we designed synthetic oligomers targeting the c-isoforms of fibroblast growth factor receptors (FGFRs), which induced robust MAP kinase pathway activation, sustained calcium signaling, and, in an iPSC-to-endothelial cell differentiation model, promoted arterial cell fate. In contrast, native FGF, which engages both b- and c-isoforms of the FGFR family, primarily directed cells toward a venous identity in the same system.

In a second study, we applied de novo protein design to develop agonists for the TrkA receptor, whose native ligand is nerve growth factor (NGF). By fine-tuning the spatial geometry in which two TrkA receptors are brought together, we were able to modulate signaling outcomes with high precision. The resulting NGF mimetics retained neurotrophic effects in differentiated neurons while significantly reducing nociceptive signaling, which is typically induced by native NGF in sensory neurons during wound healing. This highlights the capacity of AI-guided protein design not only to mimic but also to reprogram natural signaling functions by encoding signaling bias directly into the molecular architecture of the agonists.

We propose that integrating such de novo designed synthetic growth factor mimetics into stem-cell differentiation workflows will significantly enhance control over cell fate decisions, unlocking new opportunities in regenerative medicine, tissue engineering, and disease modeling.

Edman, Phal*, Redler*, Schlichthaerle*et al., Modulation of FGF pathway signaling and vascular differentiation using designed oligomeric assemblies. Cell, 187 (14), 3726–3740.e43 (2024). DOI: 10.1016/j.cell.2024.05.025*

Schlichthaerle, Yang*, Detraux*, Johnson*et al., Designed NGF mimetics with reduced nociceptive signatures in neurons. bioRxiv. (2025) DOI: 10.1101/2025.04.14.648806.*

Abstract No. T13

Enhanced fibrosis modeling in human skin organoids through hiPSC-derived macrophage integration

Amelie Reigl[1,*], *Anna-Sophie Hauser*[1,2], *Shanice Gundel*[2], *Nina Köder*[1], *Saskia Zöphel*[3], *Pauline Klose*[1,2], *Johanna Scherer*[1,2], *Maximilian Schinke*[4], *Mania Ackermann*[4], *Florian Groeber-Becke*[1,5], *Nico Lachmann*[4], and *Dieter Groneberg*[1]

[1] Fraunhofer ISC Translational Center Regenerative Therapies, Würzburg
[2] Julius-Maximilians University Würzburg
[3] University Hospital Würzburg
[4] Fraunhofer Institute for Toxicology and Experimental Medicine (ITEM), Hannover
[5] University Hospital of Düsseldorf, Experimental Ophthalmology

*Presenting author

Animal models are limited in translational value due to fundamental differences between murine and human skin. To address these issues, we generated a complex three-dimensional skin organoids (SO) from human induced pluripotent stem cells (hiPSCs), based on a modified protocol by Lee et al. The SO replicate all three skin layers, including hair formation. We characterized the SO using molecular markers for the epidermal layer (CK5, CK17), dermal layer (vimentin), and hypodermal layer with adipocytes using lipid staining (Nile Red).

To model fibrotic remodeling, we stimulated mature SO with TGF-β for seven days and analyzed tissue responses via immunofluorescence for α-SMA, FAP, collagen I, and Ki67. To capture immune tissue interactions, we integrated RFP-labeled hiPSC-derived macrophages (provided by the group of Prof. Nico Lachmann) into the SO and maintain long-term co-culture. During the incubation we monitored the effects of macrophages on the composition of the extracellular matrix, tissue architecture, and cell proliferation. Significant transcriptional differences in fibrosis-associated markers (e.g. ACTA2, FAP) were observed by qPCR with and without the presence of macrophages indicated by both conditions formation of myofibroblast. In the condition with macrophages altered Ki67 and reduced expression of Col1A1 were observed suggesting a modulating role of immune cells during fibrotic remodeling. We treated the organoids with the antifibrotic drug nintedanib, which attenuated the fibrotic phenotype.

Our model enables the functional testing of fibrosis-inducing stimuli and immune interactions in a translational human test system. This provides an alternative to animal models for the study of fibrotic skin conditions and integration of macrophages led to a more translational in vitro system.

Abstract No. T14

Heart organoids and engineered heart tissue as hiPSC-derived in vitro models for Fabry cardiomyopathy

Anna Reinelt[1,*], *Sandra Laufer*[1], *Linda Blomberg*[2], *Christine E. Kurschat*[2], *Nicola Wanner*[1], *Nicole Muschol*[3], *Annika Gathmann*[1], *Michaela Schweizer*[4], *Nikolay Zolotarev*[1], *Arne Hansen*[5], *Tobias B. Huber*[1], and *Fabian Braun*[1,7]

[1] Hamburg Center for Kidney Health (HCKH) & III. Dep. Medicine, Uni. Med. Center Hamburg-Eppendorf
[2] Center for Mol. Med. Cologne, Center for Rare Diseases & Dep. II Intern. Med., Uni. Cologne
[3] Dep. of Pediatrics, Uni. Med. Center Hamburg-Eppendorf
[4] Center for Mol. Neurobiol. Hamburg (ZMNH), Uni. Med. Center Hamburg-Eppendorf
[5] Dep. of Exp. Pharmacol. and Toxicol., Uni. Med. Center Hamburg-Eppendorf
[7] Martin Zeitz Center for Rare Diseases**,** Uni. Med. Center Hamburg-Eppendorf

*Presenting author

Background: Accurate models of Fabry cardiomyopathy are currently lacking in conventional rodent systems. Therefore, our project aims to develop advanced human *in vitro* systems to improve our molecular understanding of the cardiac phenotype in Fabry disease. To create these informative systems, we use human induced pluripotent stem cells (hiPSCs) derived from Fabry patients and differentiate them into heart organoids and engineered heart tissues (EHTs).

Methods: We reprogrammed hiPSCs from primary urinary cells (PUCs) collected from Fabry patients with various pathogenic variants. Isogenic control lines were created through CRISPR/Cas9 gene editing. Using published protocols, we differentiated these cell lines into heart organoids as well as cardiomyocytes for the generation of EHTs.

Results: We validated the loss of alpha-galactosidase A protein, deficiency of alpha-galactosidase A enzyme activity and the accumulation of globotriaosylceramide (Gb3) in Fabry heart organoids and EHTs. Additionally, targeted metabolomics analysis confirmed the presence of Gb3 subspecies in diseased heart organoids. This pathophysiology did not impair the differentiation. Heart organoids showed robust contractions and snRNA-seq analyses exhibited several cardiac cell types, including cardiomyocytes, characterized by sarcomeres and Z-disc formation which we could also observe in electron microscopy. We implemented treatment with enzyme replacement therapy and, where applicable, chaperone therapy. Fabry EHTs were prone to arrhythmias, reflecting the clinical phenotype of Fabry patients. Administration of enzyme replacement therapy reduced substrate accumulation but did not alter the functional impairment.

Conclusion: We have established these novel, unique platforms for the investigation of human Fabry cardiomyopathy using *in vitro* disease models. Current analyses are focused on identifying differential genes expression from the snRNA-seq dataset which will then be complemented with deep proteomic and spatial phenotyping combined with the administration of available therapies.

***Support**: Amicus Therapeutics, Federal Ministry of Research Technology and Space*

Abstract No. T15

iPSC-driven drug discovery identifies sildenafil as a drug candidate for the mitochondrial disease Leigh syndrome

Annika Zink[1,*], *Dao-Fu Dai*[2], *Annika Wittich*[3], *Giulia Pedrotti*[4], *Sonja Heiduschka*[1], *Guillem Aguilar*[5], *Sofia Notopoulou*[6], *Tancredi Massimo Pentimalli*[7], *Spyros Petrakis*[6], *Anu Soumalainen*[8], *Ole Pless*[3], *Emanuela Bottani*[4], *Antonio del Sol*[5], *Markus Schuelke*[9], and *Alessandro Prigione*[1]

[1] University Hospital Düsseldorf, Heinrich Heine University
[2] Johns Hopkins University School of Medicine, Baltimore, US
[3] Fraunhofer Institute for Translational Medicine and Pharmacology ITMP, Hamburg
[4] University of Verona, Italy
[5] University of Luxembourg
[6] Institute of Applied Biosciences (INAB), Thessaloniki, Greece
[7] Max Delbrück Center for Molecular Medicine in the Helmholtz Association (MDC), Berlin
[8] University of Helsinki, Finland
[9] Charité - Universitätsmedizin Berlin

*Presenting author

Leigh syndrome (LS) is a severe and rare genetic form of mitochondrial disease that causes psychomotor regression and acute metabolic crises. Most individuals with LS have defects in mitochondrial energy production, resulting from pathogenic gene variants in over 100 genes in the nuclear DNA (e.g. *SURF1*) or mitochondrial DNA (e.g. *MT-ATP6*), involved in oxidative phosphorylation (OXPHOS). To accelerate drug discovery for LS, we employed a drug repurposing approach using induced pluripotent stem cell (iPSC)-derived neural progenitor cells (NPCs) from patients with LS to screen a library of 5,632 compounds that are potentially repurposable. We identified phosphodiesterase 5 inhibitors (PDE5i) as leads capable of normalizing mitochondrial membrane potential in LS NPCs. Among PDE5i, we prioritized sildenafil due to its safety profile in children and adults. Sildenafil was shown to restore key signaling pathways involved in nervous system development, promote neurite outgrowth in LS neurons, and normalize aberrant calcium responses in LS brain organoids under metabolic stress. Sildenafil extended lifespan in a mouse model of LS and rescued both metabolic and neurological phenotypes. Furthermore, sildenafil showed acceptable blood-brain barrier permeability, as demonstrated in an iPSC-derived model. Notably, chronic off-label compassionate treatment with sildenafil in six LS patients was well tolerated and associated with improved motor function and increased resilience to metabolic crises. These findings underscore the promise of iPSC-based drug discovery for rare, currently untreatable disorders and support sildenafil as a potential therapeutic candidate for LS.

Abstract No. T16

Blood stem cell expansion: Technology to biology

Adam Wilkinson

University of Cambridge & Cambridge Stem Cell Institute, UK

Haematopoietic stem cells (HSCs) are a rare but potent cell type that support life-long haematopoiesis and stably regenerate the blood and immune system following transplantation. HSC transplantation represents a mainstay treatment for various diseases of the blood and immune systems. The ex vivo expansion and manipulation of HSCs therefore represents an important approach to ask biological questions in experimental haematology and to help improve clinical HSC transplantation therapies. However, for many years it remained challenging to expand transplantable HSCs ex vivo. I will summarise our recent improvements in expanding functional HSCs ex vivo using polymer-based culture conditions. I will also provide examples of the biomedical applications of this technology, including how we can use these systems to uncover new biological mechanisms regulating HSC activity.

Abstract No. T17

C57BL/6 Kit-mutant immunodeficient mouse models uncover mechanism of successful hematopoietic stem cell xenotransplantation

Julia Fröbel[1,*], *Susann Rahmig*[1,2], *Emilie Coppin*[1,2], *Jonas Metz*[3], *Thomas Höfer*[3], and *Claudia Waskow*[1,4]

[1] Leibniz Institute on Aging – Fritz Lipmann Institute (FLI), Jena
[2] Institute of Immunology, Technical University Dresden
[3] Deutsches Krebsforschungszentrum (DKFZ), Heidelberg
[4] Friedrich-Schiller University Jena

*Presenting author

A proper bone marrow microenvironment is a key factor for hematopoietic stem cells, endogenous as well as transplanted, supporting their engraftment, maintenance and differentiation. Xenotransplantation models are limited due to host immune system rejection and accessibility of the niche space; challenges that were addressed by immunodeficient mice and irradiation of the recipients to deplete endogenous HSCs. But irradiation also leads to negative side effects on the bone marrow niche causing an inflammatory environment for the incoming HSCs. Introducing a *Kit*-deficiency into immunodeficient recipient mice, we have overcome this need for irradiation enabling HSC engraftment into a natural microenvironment. In these non-irradiated recipients, human donor HSCs show robust long-term engraftment and multi-lineage hematopoietic output of human immune cells. The niche responds to donor cells by specific increase of mesenchymal stroma cell (MSC) numbers and overall adjust transcriptional and functional characteristics mirroring long-term post-HSCT symptoms in patients. To determine the specific niche cell type and molecular signalling axis important for donor-HSC-supporting function, we established a modular xenotransplantation mouse model on C57BL/6 background allowing the introduction of further genetic modifications. We here show that human HSC engraftment in mice depends on Lepr$^+$ MSCs and that the SCF-Kit signalling axis between murine Lepr$^+$ MSCs and human HSCs is a key molecular factor allowing cellular and molecular interspecies-crosstalk mediating engraftment and maintenance of xenogenic hematopoietic stem cells in mice.

Abstract No. T18

From codons to blood: A new perspective on hematopoietic differentiation

Marius Külp[1,*], *Adrien Jolly*[1], *Tessa Schmachtel*[1], *Björn Häupl*[1], *Thomas Oellerich*[1], *Halvard Bönig*[2], and *Michael Rieger*[1]

[1] Department of Medicine 2 - Hematology and Oncology, Goethe University Frankfurt am Main
[2] German Red Cross Blood Service Baden-Württemberg-Hessen, Frankfurt am Main

*Presenting author

The genetic code governs the structure and function of RNA and proteins. Although all cells use the same 64 codons, cells differ in their transcriptomic codon usage and transfer RNA (tRNA) expression. We established a precision approach to track the genetic code from genome to proteome, integrating mRNA and tRNA transcriptomes, the translatome and the proteome of cell populations at different stages of hematopoiesis to deconstruct hematopoietic differentiation at single codon resolution.

Development of a low-input tRNA-seq approach enabled us to explore the tRNA expression profiles of sorted hematopoietic stem cells (HSCs), progenitors and T cells derived from healthy human donors. Importantly, tRNA expression was sufficient to separate HSPCs and T cells in an unsupervised clustering analysis, indicating contribution of tRNA profiles to cellular identities. Moreover, we identified 25 differentially expressed tRNAs.

Upon integration of tRNA demand, generated by the transcriptomic codon usage, with tRNA supply, represented by tRNA expression, we identified lowly and highly supplied codons. For distinct codons, the relative tRNA supply changed along the hematopoietic hierarchy. Their impact on translational fidelity is currently analyzed by alignment with translatomes and proteomes using machine learning and perturbation experiments.

We validated our approach with peripheral blood of 10 healthy donors, and confirmed codon-specific supply levels. Using a meta-ORF-analysis, we found an enrichment of highly supplied, and a depletion of lowly supplied codons at translation start sites, pointing to a regulation of protein abundance by codon positioning in human blood cells.

In conclusion, we describe differential tRNA expression within the hematopoietic system, track codons from genome to proteome, and dissect mechanisms of translational control potentially driving cell states.

Abstract No. T19

Multimodal omics identify an age resilient multipotent progenitor population that preserves youthful identity and function in the hematopoietic system

Mari Carmen Romero-Mulero[1, 2, *], *Julian Mess*[2], *Mirijam Egg*[2], *Maria-Eleni Lalioti*[1], *Yu Wei Zhang*[2], *Jasmin Rettkowski*[1], *Helen Demollin*[2], *Lucie Hustin*[3], *Alessandro Donada*[3], *Karin Jäcklein*[2], *Beatriz Silva-Rego*[2], *Carolin Wadle*[1], *Francesco Cardamone*[2], *Isabella M. Mayer*[1], *Dawn S Lin*[4], *Pia Sommerkamp*[4], *Katharina Schönberger*[2], *Leila Perié*[3], *Andreas Trumpp*[4], *Marta Derecka*[5], *Eirini Trompouki*[6], and *Nina Cabezas-Wallscheid*[1]

[1] ETH Zürich, Switzerland
[2] Max Planck Institute of Immunobiology and Epigenetics, Freiburg
[3] Institut Curie, Paris, France
[4] German Cancer Research Center (DKFZ), Heidelberg
[5] St. Jude Children's Research Hospital, Memphis, US
[6] IRCAN Institute for Research on Cancer and Aging, Nice, France

*Presenting author

Hematopoietic stem cells (HSCs) and multipotent progenitors (MPPs) play critical roles in maintaining hematopoietic homeostasis. Although HSC aging has been a field of major interest, the molecular and functional changes across early progenitor subsets spanning lifetime remain poorly understood. Here we provide a comprehensive multiomic and functional characterization of HSCs and MPP1–6 populations in young and aged mice. Using bulk RNA-seq, single cell 10x Multiome (scRNA-seq and scATAC-seq), metabolomics, lipidomics and functional assays, we identify unique features and regulatory mechanisms that maintain the role and identity of hematopoietic stem and progenitor cell (HSPC) subsets in the hematopoietic system. We capture widespread age-associated alterations in these populations, particularly HSCs, including chromatin remodeling, metabolic imbalance, and functional decline. Strikingly, the CD150^{neg} MPP6 emerges as a uniquely preserved population with age, retaining a youthful molecular profile and sustained capacity for long-term lymphoid reconstitution. Our findings refine the model of the hematopoietic hierarchy in aging and establish MPP6 as a resilient population with potential relevance for therapeutic strategies aimed at restoring balanced hematopoiesis in the elderly.

Romero-Mulero, M.C., Mess*, J., Egg*, M., Lalioti, et al. Multimodal omics identify an age resilient multipotent progenitor population that preserves youthful identity and function in the hematopoietic system. Manuscript under revision.*

Abstract No. T20

Development of accessible ex vivo culture models of the human hematopoietic bone marrow niche

Sigrid Uxa[1,*], *Amanpreet Kaur Bains*[1], *Donata Backhaus*[1], *Sophia Schmidt*[1], *Paula Sophie Reichelt*[1], *Bruno Ederer*[1], *Marie Schneider*[1], *Uwe Platzbecker*[2], and *Michael Cross*[1]

[1] University Leipzig
[2] University Clinic Leipzig

*Presenting author

Leukemogenesis, progressing from clonal hematopoiesis (CH) through myelodysplastic neoplasia (MDS) to acute myeloid leukemia (AML), originates in a hematopoietic stem cell through a complex interplay of genetic mutations, epigenetic alterations, and bone marrow inflammation, all occurring within a specialized hematopoietic niche. Our ongoing efforts to understand this progression and to develop effective, targeted treatments rely heavily on advanced culture models that faithfully replicate the microenvironment of human bone marrow.

To overcome the limitations of standard monoculture, adherent long-term bone marrow culture (LTBMC), and animal models, we have established a three-dimensional (3D) suspended aggregate culture system. This combines patient-derived CD34+ hematopoietic stem and progenitor cells (HSPC) with pre-cultured stromal components, including mesenchymal, endothelial, and osteoprogenitor cells. Micro-aggregates, each containing a few hundred stromal and hematopoietic cells, are first formed in 5D Kugelmeiers plates using a carefully optimized medium that supports both compartments equally. After four days, these micro-aggregates are pooled into multiple wells of a U-bottomed, ultra-low adhesion 96-well plate, where they merge into larger aggregates and start producing myeloid progeny within a week. Maturing cells detach passively and accumulate at the base of the well, minimizing disturbance to the niche and allowing for repeated sampling over several weeks. Hematopoietic activity is sustained for at least five to six weeks, often much longer. The addition of erythropoietin to the culture stimulates robust erythropoiesis.

Compared to conventional adherent systems, this aggregate culture offers multiple advantages: small scale, high reproducibility, rapid onset of active hematopoiesis, and reliable support for all CH, MDS, and AML patient samples tested, confirmed by retention of disease-specific markers. These unique features make the system well-suited for investigating niche biology and for personalized preclinical drug screening. We are currently studying patient-specific drug responses in MDS, refining erythropoiesis conditions, and exploring metabolic differences between erythropoiesis and granulopoiesis.

Abstract No. T21

Deep immune-phenotyping of HLA-homozygous iPS-cardiomyocytes

Dirk Strunk

Paracelsus Medical University, Salzburg, Austria

Immunogenicity of donor-derived human induced pluripotent stem cell (hiPSC)-derived transplants limits their applicability in regenerative medicine. Selecting human leukocyte antigen-homozygous hiPSC progeny for cell transplantation could be a mitigation strategy and haplo-identical matching would expand the number of recipients. Here we asked whether deep-immune phenotyping of hiPSC-derived cardiomyocytes (iPS-CM) can predict their immunogenicity.

We differentiated iPS-CM from hiPSC of two HLA-homozygous donors and two HLA-heterozygous controls in 3D cultures under chemically defined conditions into iPS-CM. Broad immune-phenotype screening of 354 target molecules revealed differential expression of 101 molecules between iPS-CM and parental hiPSC. We selected 54 markers for deep immune-phenotyping by spectral flow cytometry. Dimensionality reduction clearly discriminated iPS-CM from hiPSC and enabled monitoring interferon-gamma (IFN-γ)-responsive phenotype patterns as a surrogate of immune-reactivity. Compared to heterozygous counterparts HLA-homozygous iPS-CM displayed significantly reduced HLA-ABC levels with otherwise comparable immune-phenotype. IFN- γ-stimulated iPS-CM upregulated HLA-ABC, HLA-E, PD-L1 and the don't-eat-me signal CD47 arguing towards favorable immune recognition profile of iPS-CM under inflammatory/stressed conditions.

Abstract No. T22

Biological-engineering and application of a myocardial patch

Joseph Petrus Gerardus Sluijter

University Medical Center (UMC) Utrecht, The Netherlands

Myocardial infarction (MI) leads to irreversible loss of cardiomyocytes and replacement by non-contractile scar tissue, ultimately culminating in adverse remodeling and heart failure. To address this, our recent work explores the engineering of a biomimetic myocardial patch capable of structural, electrical, and functional integration with host tissue.

We use human pluripotent stem cell–derived cardiomyocytes, endothelial cells, , and fibroblasts to recapitulate native myocardial cellular complexity. Protocols are developed for iPSC-CM expansion and differentiation, whereas scaffold fabrication employs three-dimensional (3D) bioprinting of biomaterials that replicate the architecture and mechanical anisotropy of the native extracellular matrix (ECM) to further support cell alignment, electrical coupling, and mature contractile behavior.

The engineered patch demonstrates high viability, synchronous beating, and enhanced cell coupling in vitro, while preclinical in vivo application of the patch to infarcted myocardium resulted in ventricular improvements, incl enhanced neovascularization and reduced fibrotic remodeling. Our integrated strategy combining stem-cell derived cardiac cell types, biomimetic 3D printed scaffolds, and multifunctionalization renders a cardiac patch with potent reparative capacity.

Abstract No. T23

Large-scale production of human pluripotent stem cell–derived cardiomyocytes

*Kevin Cyrys**, *Nils Kriedemann*, *Carlos A. Hernandez Bautista*, *Jana Teske*, *Annika Franke*, *Ulrich Martin*, and *Robert Zweigerdt*

Lebao, MHH, Hannover

*Presenting author

Human pluripotent stem cell (hPSCs)-derived cardiomyocytes (hPSC-CMs) have great potential for regenerative medicine, including preclinical studies in large animal models and tissue engineering. However, robust and economically viable production processes compatible with industry and regulatory standards are required. Given the limited proliferation of hPSC-CMs, pre-expansion of hPSCs to clinically relevant process scales is mandatory prior to their directed cardiomyogenic differentiation.

Here we present a full production pipeline in suspension culture (3D) in stirred tank bioreactors (STBRs), inoculated either with hPSCs derived from adherent (2D) pre-culture (termed: “conventional inoculation”) or directly from cryopreserved “hPSC high-density seeds” (HDS; termed: “direct inoculation”). The process is successively scaled up to 2,000 mL culture scale via a seed train approach. For cell passaging at each upscaling step, EDTA-based dissociation of the suspension culture-induced hPSC aggregates directly in the bioreactor vessel was enabled, while ensuring maintenance of pluripotency and exponential proliferation kinetics.

Finally, cardiomyogenic differentiation in a chemically defined, protein-free medium formulation in 2,000 mL culture scale was performed, yielding >1.3 × 109 cardiomyocytes with typical lineage purity of >90%.

The work demonstrates the robust, scalable production of human cardiomyocytes in clinically relevant quantity and quality while substantially reducing media costs, providing an important step towards GMP-compliant cell production and therapeutic application.

Abstract No. T24

Automated myocardial cell injection

Raphael Mönkemöller

Institute for Mechatronic Systems, Leibniz University Hannover

Introduction

Intramyocardial cell therapy offers a promising approach to mitigate the long-term effects of heart failure. However, physiological tremor, very small injection volumes (<50 µL) and especially the dynamic heart motion make manual transepicardial injection a challenging process. These factors often result in cell loss caused by myocardial perforation, backflow, and low retention rates. To address these challenges, we propose a novel robotic system to automate and enhance the injection process. The system comprises three integrated subsystems: a precision injector head, a positioning mechanism, and a cardiac tissue stabilizer.

Methods

The stabilizer, similar to conventional cardiac vacuum stabilizers, reduces the heart wall motion and is able to extend its suction feet to enhance the motion reduction via stretching and enables use through smaller incisions (2cm x 2cm). The injector head delivers very small volumes with high accuracy and repeatability. It achieves precise injection depth using a needle sheathing with vacuum suction acting as a stamp and suction cup. The positioning system tracks residual cardiac motion via applied surgical markers and a stereo camera. It positions the injection head using a delta kinematic for fine and a surgical assistant arm for rough positioning.

Results

The stabilizer reduces motion amplitude by 75%, comparable to state-of-the-art devices. The dosage mechanism is able to achieve a accuracy below 5% error while the injector with the needle sheathing achieves submillimeter depth accuracy. In addition, especially the stabilizer and the injector subsystems can be used on their own to improve the cell injection process and to improve injection accuracy and repeatability in other settings.

Conclusion

The device achieves a significant improvement in terms of accuracy and precision. This might help the cells to achieve better retention and increase their survivability. Future work will focus on preventing cell sedimentation, miniaturizing the applicator and conducting in-vivo trials.

Abstract No. T25

Developing a safe and immune-evasive stem cell source for off-the-shelf therapeutic products

Andras Nagy

Sinai Health System, Toronto, Canada

Ex-vivo cultured allergenic cells are currently being evaluated for treating degenerative conditions. However, achieving allograft acceptance without immune suppression is one of the greatest challenges. To tackle this issue, with transgenically expressed eight local acting immune modulatory transgene in the cells. We hypothesized that this would protect them against rejection and attain allogeneic acceptance ("AlloAccept") in fully immune-competent mice. Our results indicated that allogeneic allograft survived the long-term without the use of immunosuppressive drugs.

Since immune-modulatory genes have highly conserved functions, this strategy could also be effective in humans. By utilizing differentiated derivatives of human "AlloAccept" embryonic stem cells in humanized immune system mice, we demonstrated their long-term persistence in allogeneic settings and therapeutic effects for conditions, such as Parkinson's disease, arthritis and diabetes. Therefore, the "AlloAccept" system could provide off-the-shelf cells to treat or cure medical conditions without requiring the patients' immune suppression.

Ensuring the safety of immune-evasive cell-based therapies is critical, as these cells bypass natural immune surveillance and could pose a tumorigenic risk. To address this, we developed "SafeCell"—a highly reliable kill-switch system designed to eliminate cells exhibiting cancerous potential or uncontrolled proliferation.

Beyond enabling rapid clearance of undesired cells, "SafeCell' generates safety data, empowering clinicians, regulators, and patients to make informed decisions. This genome edit system paves the way for safer and more effective cell therapies, with the potential to transform modern medicine.

The "SafeCell" system, when combined with "AlloAccept" genome editing, creates a single pluripotent cell line that serves as a source of readily available therapeutic cell products. These cells can be further modified to express biologics to improve their therapeutic potential. Some models will be presented for such combination of gene and allogeneic cell therapies.

Abstract No. T26

Lineage single-cell multi-omics identify novel regulators and barriers in iPSC reprogramming

Angeliki Spathopoulou[1,*], *Niklas Schweiger*[1], *Katharina Günther*[1], *Esk Christopher*[1], *Francesca Finotello*[1,2], and *Frank Edenhofer*[1]

[1] Molecular biology & CMBI, University of Innsbruck, Austria
[2] Digital Science Center (DiSC), University of Innsbruck, Austria

*Presenting author

The induction of pluripotency from somatic cells through the ectopic expression of OCT4, SOX2, KLF4, and cMYC is a cornerstone in biomedical research and regenerative medicine, yet the molecular dynamics governing successful reprogramming remain partially understood. To address this, we applied a multimodal single-cell approach to dissect the transcriptional and epigenetic trajectories of induced pluripotent stem cell (iPSC) reprogramming. By integrating single-nucleus RNA and ATAC sequencing data with lentiviral barcoding for lineage tracing, we profiled four key timepoints (day 0, 7, 13, and 20) to reconstruct cell state transitions during reprogramming. Lineage analysis revealed that only a minority of cells reprogrammed successfully, while the majority diverged toward non-pluripotent fates, uncovering critical bifurcation points that restrict complete reprogramming. Temporal transcriptome dynamics identified intermediate populations marked by progressive loss of somatic identity (e.g., *COL1A1*) and gradual acquisition of pluripotency (e.g., *LIN28A*/*LIN28B*), revealing a continuum of transcriptional states. Notably, our analysis identifies a previously uncharacterized transcription factor that appears to enhance the mesenchymal-to-epithelial transition (MET), a critical step in early reprogramming. While our functional validation studies are ongoing, this factor presents a promising candidate for molecular understanding of reprogramming mechanisms. Our integrative, lineage-tracing multi-omics approach reveals new regulators in cellular plasticity and provides a tool for enhancing stable iPSC reprogramming. These findings offer novel insights into the transcriptional control of cellular plasticity and reprogramming and hold potential for improving regenerative strategies in basic and translational settings.

Abstract No. T27

You shall not pass - functional pluripotency testing using hiPSCore

Jochen Dobner[1,*], *Sebastian Diecke*[2], *Jean Krutmann*[1], *Alessandro Prigione*[3], and *Andrea Rossi*[1]

[1] IUF - Leibniz research institute for environmental medicine, Düsseldorf
[2] Max Delbrück Center for Molecular Medicine, Berlin
[3] University Hospital, Heinrich-Heine-University, Düsseldorf

*Presenting author

Background: Functional pluripotency testing ensures high quality of hiPSC starting material for sophisticated differentiation experiments, but methods for the assessment are, despite some efforts from the community, still poorly standardized. In addition, their application is often based on subjective evaluation rather than reliable automated assessment methods.

Methods & Results: In the context of directed trilineage differentiation, we have recently identified a set of 172 marker genes not covered by current guidelines (Dobner et al, 2024). We have identified and thoroughly validated a core set of marker genes that are useful to discriminate early PSC cell fates analyzing more than 25 hiPSC lines. These lines include commercially available cells, reprogrammed cells from skin and urine cells, genome-edited cell lines, and hESC lines. To enable, automated, unbiased, and reliable assessment of undifferentiated and early germ layer-differentiated PSCs, we have developed a machine learning-based qPCR assay and classification software: *hiPSCore* enables fast, easy, and accurate assessment of early PSC cell fates and functional pluripotency testing. We further demonstrate that recurrent testing for cell fates allows accurate and convenient assessment of the quality of running cultures enabling cost- and time-effective planning and execution of long downstream differentiation protocols. Further, we are currently developing a resource to understand early cell fate decision by validating more than 50 additional genes derived from our published RNAseq dataset. A comprehensible database to assess this expression data in the context of transcriptomic datasets is currently in preparation.

Conclusion: Functional and recurrent pluripotency testing using hiPSCore is not just a fun way to spend time in the lab, but rather an efficient means of saving both precious time and reagents in the context of hPSC-related differentiation experiments.

Dobner, J., Diecke, S., Krutmann, J. et al. Reassessment of marker genes in human induced pluripotent stem cells for enhanced quality control. Nat Commun 15, 8547 (2024). https://doi.org/10.1038/s41467-024-52922-1

Abstract No. T28

PluripotencyScreen – quantification of early cell fate decisions of iPSC based on DNA methylation

Kira Zeevaert[1,*], *Vera Nischalke*[1], *On-Chung Ian Shum*[1], *René Krüger*[1], *Marco Schmidt*[1], *Mohamed H. Elsafi Mabrouk*[1], *Lena Hahnfeld*[1], *Tom Börstler*[2], *Beate Winner*[2], and *Wolfgang Wagner*[1]

[1] Institute of Stem Cell Biology, Helmholtz-Institute for Biomedical Engineering, RWTH Aachen University Medical School
[2] Department of Stem Cell Biology, Friedrich-Alexander Universität Erlangen-Nürnberg (FAU)

*Presenting author

Quality control of induced pluripotent stem cells (iPSC) remains a challenge. To validate pluripotency, the trilineage differentiation potential towards endoderm, mesoderm, and ectoderm is crucial. Conventional quality control assays involve assessing morphological criteria, surface markers, gene expression changes, or teratoma formation in mouse models, but these are difficult to standardize and quantify. Recognizing the increasing need to accurately characterize iPSC and their derivates, we established PluripotencyScreen – an epigenetic biomarker to track early cell fate decisions through DNA methylation at specific CG dinucleotides (CpG). Using the computational framework CimpleG, we identified three characteristic CpG for the pluripotent state and the individual differentiation states (endoderm, mesoderm, and ectoderm) based on our own Illumina Methylation BeadChip data. From these signatures, we established a pluripotency score, that not only tracks reprogramming of iPSC but also indicates differentiation efficiency. Our lineage-specific scores monitor both, the directed differentiation of iPSC and multilineage undirected differentiation in embryoid bodies. The assay was able to reveal iPSC lines with inhibited ectodermal differentiation efficiency in embryoid bodies and reflected the impact of culture media and differences between directed and undirected differentiation towards neuroectoderm. To improve efficiency, we have optimized sample shipping on Protein Saver Cards, where cells only need to be dried onto, DNA isolation and bisulfite conversion protocols, and established digital PCR assays, which allow targeted measurement of several CpG simultaneously. Currently, we are validating an updated signature covering a wide range of differentiation protocols and cell lines. PluripotencyScreen aims to be a reliable and reproducible quality control of iPSC by quantifying pluripotency and early lineage-specific commitment.

Schmidt M, Zeevaert K, Elsafi Mabrouk MH, et al. 2023. Epigenetic biomarkers to track differentiation of pluripotent stem cells. Stem Cell Reports 18: 145-58.

Abstract No. T29

Does gene dosage influence cell fate during human gastrulation?

Theresa Gross-Thebing[1,*], *Geraldine M Jowett*[1], *Jitesh Neupane*[1], *Frederick CK Wong*[1], *Nelly Drobjazko*[1], *Alexander Karpovitsch*[1], *Mien Chew*[1], *Richard Butler*[1], *Sargon Gross-Thebing*[1], *Anastasia Sybirna*[1], *Liisa van Vliet*[2], *Vasily Kantsler*[3], *Wolfram H Gruhn*[3], *Emma L Rawlins*[1], *Florian Hollfelder*[2], *Ramiro Alberio*[4], and *Azim Surani*[1]

[1] Gurdon Institute, University of Cambridge, UK
[2] Department of Biochemistry, University of Cambridge, UK
[3] University of Warwick, UK
[4] University of Nottingham, UK

*Presenting author

Transcription factors are essential regulators of early human development, but their dosage relevance remains unclear. In humans, the transcription factor SOX17 acts combinatorially with PRDM1 to specify germ cell and endoderm lineages during gastrulation, where imbalances can lead to incomplete germline induction or aberrant endodermal differentiation. This suggests that maintaining a precise balance of SOX17-PRDM1 is probably vital for cell fate determination during early embryonic development. Using a protein degron system in human embryonic stem cells, I established tunable control of SOX17 protein levels to investigate the downstream effects on early lineage specification. During differentiation to definitive endoderm, SOX17 titration revealed a dose-dependent response in the expression of target genes, including PRDM1 and FOXA2. These results suggest that SOX17 functions in a graded manner to modulate transcriptional networks underpinning early differentiation. To study these dynamics in a physiological context, I implemented a 2D-micropatterned gastruloid model that enhances differentiation of germline and endodermal lineages and enables live-cell imaging, aiming to assess the impact of transcription factor dosage on cell fate determination. In parallel, I am investigating whether SOX17 dosage affects development *in vivo*, by examining human embryonic tissue and porcine embryos. While findings of this study emphasize the sensitivity of developmental programs to transcription factor dosage, this integrated approach provides a foundation for future studies into how quantitative regulation of transcription factors contributes to robust yet flexible lineage specification in early human development. The resulting mechanistic insight will enhance our understanding of dosage-sensitive diseases and inform applications in regenerative medicine.

Abstract No. T30

Modulating reactive astrocytes in Alzheimer's disease[#]

Henrik Renner, *Stefanie Maedje*, *Saskia Stoltze*, *Amodio Luca Balzano*, *Alfonso Gerardo Garcia Diaz Barriga*, *Alberto Perez Alvarez*, *Malte Simon*, *Carina Wollnik*, *Rachel Mangan*, *Claudia Ploetzky*, *Nico Michaelsen*, *Marco Delmonte*, *Melanie Carquin*, *Romain Chassefeyre*, *Joachim Ahlbeck*, *Kai Funke*, *Alexandra Hackmann*, *Thomas Palm*, *Kenneth Young*, *Fabrizio Micheli*, *Jonathan Bentley*, *Mark Gemkow*, *Rainer Kuhn*, *Sandra Lubitz*, *Doris Gruber-Schoffnegger*, and *Bastian Zimmer*[*]

Evotec SE, Hamburg

*Presenting author

Astrocytes are the most abundant cell type in the human brain and play essential roles in both health and disease. In neurodegenerative disorders such as Alzheimer's disease (AD), astrocytes become chronically reactive, losing neurosupportive functions and acquiring neurotoxic properties that contribute to disease progression.

We analyzed large-scale single-nucleus transcriptomic data from AD patients to identify astrocyte-specific disease markers. Using human induced pluripotent stem cell (hiPSC)-derived astrocytes combined with inflammatory stimulation, we successfully recapitulated the upregulation of these markers in vitro. Gene knockdown and antibody-blocking approaches validated the role of a key marker in driving the reactive phenotype.

To discover small molecules that modulate this reactive state, we performed a high-throughput screen (HTS) of ~265,000 compounds using a qPCR-based readout. Compounds reducing marker expression were validated and profiled, followed by medicinal chemistry assessment to prioritize four structurally diverse chemical series.

Further evaluation including protein quantification, functional assays, and transcriptomics confirmed positive effects on astrocyte biology and biochemical pathways relevant for Alzheimers disease.

[#]*Collaborative work with Bristol Myers Squibb*

Abstract No. T31

Evaluating the potential of Bruton's tyrosine kinase inhibitors to modulate microglial inflammation to attenuate multiple sclerosis progression

Alica Blenkle[1,2,*], *Anastasia Geladaris*[1,2], and *Martin S. Weber*[1,3]

[1] Fraunhofer Institute for Translational Medicine and Pharmacology ITMP, Göttingen
[2] Institute of Neuropathology, University Medical Center, Göttingen
[3] Department of Neurology, University Medical Centre, Göttingen

*Presenting author

Multiple sclerosis (MS) is the most common demyelinating inflammatory disease of the central nervous system (CNS) causing neurological disability in millions of individuals worldwide. While significant advances have been made in the development of disease modifying therapies, mitigating disease progression remains a major challenge. This accumulation of irreversible neurological deficits is centrally mediated by CNS compartmentalized inflammation that is driven by microglia.

Since microglia have been identified as key players in mechanisms leading to disease progression and neurodegeneration, microglial states are a promising therapeutic target to attenuate the progression of MS. One therapeutic strategy to regulate microglial reactivity involves the inhibition of Bruton's tyrosine kinase (BTK), a key modulator of Fc receptor (FcR)-, Toll-like receptor- and chemokine receptor-induced pro-inflammatory microglial responses.

In this study, we used human induced pluripotent stem cell (hiPSC)-derived microglia (iMG) to evaluate the efficacy of four BTK inhibitors, evobrutinib, remibrutinib, tolebrutinib, and fenebrutinib, to modulate microglial responses in a standardized side-by-side comparison. Following FcR-dependent stimulation, the compounds influenced microglial reactivity by reducing the expression of molecules associated with antigen-presentation and/or FcR-induced chemokine secretion. Notably, evobrutinib and fenebrutinib required high concentrations to cause effects, whereas remibrutinib and tolebrutinib were effective at low physiologically relevant concentrations.

These findings highlight the potential of BTK inhibitors, particularly remibrutinib and tolebrutinib, to regulate microglial functions involved in pathogenic T cell activation and migration, offering a promising therapeutic strategy for MS progression.

Abstract No. T32

A reproducible human brain tissue model to study physiological and disease-associated microglia phenotypes

Julien Klimmt[1], *Carolina Cardoso Gonçalves*[1], *Jessica Valentina Montgomery*[2], *Stephan Müller*[3], *Severin Filser*[1], *Lars Paeger*[3], *Angelika Dannert*[1], *Sigrun Roeber*[4], *Veronica Pravata*[5], *Martina Schifferer*[3], *Judit González Gallego*[1], *Silvia Cappello*[5], *Jochen Herms*[4], *Christian Haass*[3], *Joachim L. Schultze*[2,6], *Stefan F. Lichtenthaler*[3,7], *Caterina Carraro*[2,6], and *Dominik Paquet*[1,8,*]

[1] University Hospital, LMU Munich
[2] Systems Medicine, German Center for Neurodegenerative Diseases (DZNE), Bonn
[3] German Center for Neurodegenerative Diseases (DZNE), Munich
[4] Center for Neuropathology and Prion Research, LMU Munich
[5] Dep. of Physiological Genomics, Biomedical Center (BMC), Faculty of Medicine, LMU Munich
[6] Genomics and Immunoregulation, Life & Medical Sciences (LIMES) Institute, Uni. Bonn
[7] Technical University of Munich
[8] Munich Cluster for Systems Neurology (SyNergy)

*Presenting author

Objectives

Stem-cell-based in vitro models offer promising potential to elucidate human brain cell functions and interactions under physiological and pathological conditions. However, harnessing this potential is impaired by low reproducibility, maturity, and cell-type diversity of existing models. Especially, prolonged incorporation of mature microglia and studies of neuroinflammation have proven challenging. We therefore developed a 3D cortical brain tissue model (3BTM) with high reproducibility, maturity, and viability and applied it to generate a new 3xKI model of Alzheimer's disease.

Methods

To generate 3BTMs, we applied a controlled 3-dimensional tissue engineering approach using long-term co-culture of highly pure cortical neurons, astrocytes, and microglia derived from CRISPR/Cas9-edited iPSCs.

Results

3BTMs show morphological, functional, and proteomic maturation of all cell types, leading to high similarity to their in vivo counterparts. Incorporated microglia survive for over 6 months and display mature morphology, functions, and gene expression. Importantly, when engineered to model Alzheimer's disease, 3BTMs recapitulate key disease hallmarks including amyloid deposition, increased phospho-Tau levels, and neuroinflammation, with microglia shifting their transcriptional landscape to disease-relevant signatures also found in patients. Furthermore, we could show the translational potential of the model by adding the therapeutic antibody Aducanumab, which engaged its target and promoted microglia clearance of Abeta in vitro.

Conclusions

Taken together, our model provides unprecedented possibilities to study physiological and pathological states of human brain cells, especially microglia, and it offers high applicability for developing novel therapeutic interventions for major brain diseases.

Abstract No. T33

Integration of CNTNAP2-deficient neurons into defined E/I networks unmasks functional pathophenotypes

Jasmin Aicher[1,*], *Luzia Heidrich*[1], *Natalia C. Garcia Perez*[1], *Lydia Fischer*[1], *Juan Eduardo Rodriguez-Gatica*[1], *Martin K. Schwarz*[1,2], *Lea J. Berg*[1], and *Oliver Brüstle*[1,2]

[1] University of Bonn Medical Faculty and University Hospital Bonn
[2] LIFE & BRAIN GmbH, Bonn

*Presenting author

Contactin Associated Protein 2 (CNTNAP2) deficiency is a rare disorder associated with intellectual disability, autism spectrum disorder, language impairment and epilepsy. From an evolutionary perspective, CNTNAP2 is strongly expressed in human cortex and critical for neuronal development, synaptic function, and the regulation of neuronal excitability.

We set out to explore the functional impact of CNTNAP2 in neuronal networks composed of defined fractions of forward programmed excitatory and inhibitory neurons generated by overexpression of NGN2 and ASCL1/DLX2, respectively. By systematically varying the E/I ratio, we aimed to determine how CNTNAP2 deficiency affects neuronal network function under defined circuit conditions. Excitatory NGN2$^{CNTNAP2-/-}$ neurons displayed only subtle changes, such as a decreased burst duration at 10 weeks of maturation. In contrast, networks composed of 80% NGN2$^{CNTNAP2-/-}$ and 20% inhibitory ASCL1/DLX2$^{CNTNAP2-/-}$ neurons exhibited a complex pathophenotype with a reduction in burst number, while individual bursts were longer and contained more spikes. Network burst frequency was increased overall, whereas network burst duration and spike counts were decreased.

To assess the cell-type-specific pathogenic role of CNTNAP2, we created E/I networks in which only the excitatory or the inhibitory population carried the CNTNAP2 knockout. These studies revealed that altered network function is primarily due to NGN2$^{CNTNAP2-/-}$ neurons. To further decipher underlying mechanisms, we applied AMPA, NMDA, or GABA-A receptor blockers in networks composed of 80% NGN2 and 20% ASCL1/DLX2-programmed neurons. Our data suggest that CNTNAP2 is essential for maintaining functional synaptic signalling through NMDA and GABA-A receptors. We hypothesize that CNTNAP2 orchestrates receptor trafficking dynamics, particularly of GluA1-containing AMPA receptors, thereby ensuring balanced excitatory and inhibitory transmission and proper network function. Thus, CNTNAP2 loss results in multiple, cell-type specific pathophenotypes. The data suggest that CNTNAP2 may influence receptor composition and localization at the synaptic membrane, potentially acting as a broader regulator of receptor dynamics.

Abstract No. T34

Capturing disease severity in LIS1-lissencephaly reveals proteostasis dysregulation in patient-derived forebrain organoids

Matteo Gasparotto[1,*], *Lea Zillich*[1], *Andrea Carlo Rossetti*[1], *Olivia Fechtner*[1], *Camille Maillard*[2], *Anne Hoffrichter*[1], *Eric Zillich*[1], *Ammar Jabali*[1], *Fabio Marsoner*[1], *Annasara Artioli*[1], *Ruven Wilkens*[1], *Christina Barbara Schroeter*[3], *Andreas Hentschel*[4], *Stephanie H. Witt*[1], *Nico Melzer*[3], *Sven G. Meuth*[3], *Tobias Ruck*[4], *Philipp Koch*[5], *Andreas Roos*[6], *Nadia Bahi-Buisson*[2], *Fiona Francis*[7], and *Julia Ladewig*[5]

[1] Central Institute of Mental Health, Mannheim
[2] Université Paris Descartes, Imagine Institute, France
[3] Medical Faculty and University Hospital, Heinrich Heine University Düsseldorf
[4] Ruhr University Bochum, BG University Hospital Bergmannsheil
[5] Central Institute of Mental Health, Medical Faculty Mannheim, Heidelberg University, Mannheim
[6] Centre for Neuromuscular Disorders, University Medicine Essen
[7] Sorbonne University, Paris, France

*Presenting author

LIS1-lissencephaly, caused by heterozygous LIS1 mutations, manifests as a spectrum of cortical malformations and neurodevelopmental impairment. To explore the basis of clinical heterogeneity, we generated forebrain organoids from individuals with mild, moderate, and severe forms of LIS1-lissencephaly and performed integrated cytoarchitectural, transcriptomic, and proteomic analyses.

Quantitative proteomics revealed variable LIS1 protein levels and severity-linked dysregulation of LIS1-interacting proteins, pointing to mutation-dependent functional dosage effects. Severity-correlated molecular signatures included disruption of cytoskeletal organization, adherens junction integrity, and WNT signaling. Notably, multi-omics integration uncovered progressive defects in proteostasis and endoplasmic reticulum (ER) stress, identifying unfolded protein response activation as a key mechanism associated with disease progression.

To functionally interrogate these findings, we applied everolimus, a selective mTORC1 inhibitor identified via in silico prediction, during early cortical development. Everolimus reduced premature neurogenesis and attenuated ER stress markers in moderate and severe patient-derived organoids, supporting a mechanistic link between proteostasis regulation and LIS1-related pathology.

Our findings demonstrate that multi-omics profiling of severity-stratified human organoids can capture genotype-informed gradients of dysfunction, reveal novel molecular mechanisms, and enable functional interrogation of candidate pathways in cortical malformation disorders.

Abstract No. T35

Tracing clonal architecture and differentiation trajectories in cancer using single cell barcoding

Michael Rieger

Goethe University Frankfurt, University Hospital, Department of Medicine 2 - Hematology and Oncology, Frankfurt am Main

Tumor cell heterogeneity is caused by clonal evolution induced by private somatic genetic alterations in individual tumor cells and an epigenetic tumor cell differentiation hierarchy. The models of clonal evolution and cancer stem cell (CSC) hierarchies are not mutually exclusive. Understanding the subclonal architecture and the trajectories in cancer, the advantages and challenges of clonal lineage tracing technologies including genetic barcoding will be discussed.

To track cancer architecture on clonal and single-cell resolution in vivo, we developed a lentiviral expression barcoding system in combination with scCITE-Seq in xenotransplantations of patient-derived B cell-acute lymphoblastic leukemia (ALL). While genetic heterogeneity and clonal evolution have been extensively studied, the presence of an epigenetic hierarchy is debated in ALL, and models of cell plasticity have been proposed. Intrinsic properties and local environmental influences were resolved by analyzing clonal inter-organ dissemination, including CNS infiltration.

While clonal composition was variable between recipient mice receiving the same barcoded cells, suggesting a high selective pressure by the local environment, cells with identical barcodes demonstrated consistent differentiation behavior across animals, indicating clone intrinsic differentiation properties. Single cell analysis revealed a proposed cluster of origin and distinct clusters at various stages of differentiation, which could be prospectively isolated based on markers and transplanted into NSGs. We functionally demonstrate that stages of differentiation exist with a stable population of enriched leukemia-initiating stem cells. Differential equation-based mathematical models using approximate Bayesian computation validated the most likely cluster of origin and the differentiation trajectories and excluded reversible plasticity.

The analysis of clonal ALL cells in different organs revealed a strong separation of brain infiltrating cells with distinct molecular pattern compared to the clonally related cells in other organs, with restricted clonal diversity.

Our technology provides valuable insights into the ontogeny of CSCs in ALL and will be instrumental in studying regeneration and malignancies.

Abstract No. T36

Role of aging and environmental stress in the decline of lung stem cell function and regenerative capacity

Sefa Berkay Çayir, *Sharduli Sharduli*, *Lili Rusznyak*, *Edgar Albert*, and *Maja Funk*[*]

Helmholtz Munich

*Presenting author

Background:

Organismal aging is a major driver of chronic inflammatory diseases, including Chronic Obstructive Pulmonary Disease (COPD). COPD is an incurable, age-related lung disease and a leading cause of morbidity and mortality worldwide. Its pathogenesis involves chronic inflammation of the airways, epithelial barrier dysfunction, and impaired immune responses. Aging and environmental stress disrupt lung epithelial integrity, compromising the function and regenerative capacity of lung stem and progenitor cells, thereby contributing to the onset and progression of disease.

Objective:

Understanding how aging and environmental stress impact lung epithelial stem cell function and the associated changes in cellular regeneration and immune interactions is critical for identifying early mechanisms underlying COPD and related chronic lung diseases.

Methods:

We employed 3D airway organoid models derived from young, aged, and cigarette smoke (CS)-exposed mice to investigate lung epithelial homeostasis and stem cell regenerative capacity. Organoids were analyzed for alterations in the differentiation trajectory, transcriptional profiles and DNA methylation patterns induced by aging and longitudinal CS exposure.

Results & Future Directions:

Airway organoids from aged mice exhibited diminished growth potential, indicative of stem cell exhaustion and cellular senescence. Age-associated transcriptional changes in the lung epithelium were characterized by the upregulation of inflammatory pathways, senescence markers, and cholesterol biosynthesis, with effects more pronounced than those induced by CS exposure. Ongoing studies are examining the analysis and integration of changes in the epigenetic landscapes to identify molecular candidates linked to age-dependent vulnerability and environmental injury of airway stem cells. Future work will focus on delineating the interactions between aging lung stem cells, epithelial barrier function, and immune cell crosstalk to uncover novel therapeutic targets for the prevention and treatment of COPD and other age-related pulmonary disorders.

Abstract No. T37

Investigating aging and telomere biology in lung disease using induced pluripotent stem cell derived models

Mari Ozaki[1,*], *Anja Schweikert*[1], *Fernanda Gutierrez-Rodrigues*[2], *Emma M Groarke*[2], and *Killian Hurley*[1]

[1] Royal College of Surgeons in Ireland, Dublin
[2] National Heart, Lung, and Blood Institute, National Institutes of Health, Bethesda, US

*Presenting author

Pulmonary fibrosis is a complex, age-related lung disease that leads to lung scarring, respiratory failure and death. Telomeres, repetitive DNA sequences, synthesized by telomerase protect chromosome ends from degradation. While the cause of PF is unknown, aging and short telomere length in type 2 alveolar epithelial (AT2) cells have been implicated as key players. We aimed to develop a novel iPSC derived AT2 (iAT2) model that can be used to explore aging and telomere biology in lung diseases, in particular, pulmonary fibrosis.

PBMCs from non-diseased controls were reprogrammed to iPSCs and subsequently, differentiated towards iAT2 spheroids via a directed differentiation protocol. DNA, RNA and cell pellets were collected from the PBMCs, iPSCS, at specific stages of iAT2 directed differentiation and from young and old iAT2 spheroids from non-diseased controls. Telomere length (HT-Q-FISH, Southern blot, STELA, qPCR), telomerase expression (RT-qPCR) and activity (Q-TRAP), DNA damage (FACS, IHC), proliferation (FACS, RT-qPCR), and pro-fibrotic, inflammatory and senescence gene expression (RT-qPCR) were examined to determine the aging profile of iAT2 spheroids.

Telomere length increased from PBMCs to iPSCs and then decreased throughout the different stages of iAT2 directed differentiation and aging, whilst the percentage of short telomeres increased with age. Telomere shortening was associated with decreased telomerase expression and activity, increased DNA damage and decreased proliferation. In addition, telomere shortening was associated with increased expression of senescence, anti-apoptotic and pro-fibrotic markers and decreased expression of pro-apoptotic markers.

This is the first described model of aging and telomere shortening in human iAT2 and explores the relationship of telomere length, telomerase activity, DNA damage, proliferation and pro-fibrotic pathways at specific stages of iAT2 directed differentiation. Our data supports the applicability of our model in age related lung diseases such as pulmonary fibrosis and supports the hypothesis that telomere biology plays an important role in fibrosis.

Abstract No. T38

High fat diet affects the stromal niche signaling in the porcine intestine

Falk Janos Farkas[1,]*, *Michael Sterr[1]*, *Mireia Molina van den Bosch[1]*, *Tobias Greisle[1]*, *Juliane Merl-Pham[2]*, *Christine von Törne[2]*, *Stephanie Hauck[2]*, *Yasmin Eckstein[3]*, *Simone Renner[3]*, *Eckhard Wolf[3]*, *Anika Böttcher[1]*, and *Heiko Lickert[1]*

[1] Institute of Diabetes and Regeneration Helmholtz Munich
[2] Research Unit Protein Science, Helmholtz Munich
[3] Center for Innovative Medical Models, Ludwig-Maximilians-University, Munich

*Presenting author

Diet-induced obesity is a global health burden that predisposes to type 2 diabetes and colorectal cancer. The intestine, as the body's largest endocrine organ, plays a key role in regulating glucose homeostasis and energy balance. Obesogenic diets in mice disrupt intestinal homeostasis, inducing stem cell hyperproliferation, altered differentiation, shifts in enterocyte regional identity, and enteroendocrine reprogramming. While rodent studies implicate both epithelial and subepithelial compartments in governing the intestinal stem cell (ISC) niche, the impact of a high-fat diet (HFD) on this niche remains unclear. Our initial co-culture experiments using mouse intestinal organoids and HFD-derived mesenchymal niche showed enhanced organoid formation, indicating increased niche-derived proliferative cues in HFD. However closer characterization of the subepithelial niche – especially in large animal models – is still lacking.

To address this, we employed the pig as a physiologically relevant model and analyzed small and large intestinal tissue following a 30-week HFD intervention, including both calorie-restricted and obesogenic HFD groups. Using a novel microdissection technique, we isolated ISCs together with their surrounding subepithelial niche. We then conducted integrative histological, proteomic, and single-nucleus transcriptomic profiling to assess how dietary fat alters niche signaling and epithelial differentiation.

Our data reveal reduced levels of proteins involved in metabolism and membrane transport, alongside histological signs of elevated proliferation and loss of specific enteroendocrine populations. Proteomic and transcriptomic data both show significant remodeling of the subepithelial mesenchyme in HFD-fed animals, including upregulation of VEGF, PDGF, and EGF signaling pathways, along with changes in extracellular matrix composition.

These findings indicate that dietary fat disrupts the ISC niche and impairs epithelial differentiation, independent of caloric intake. This work provides a foundational multi-modal dataset from a large animal model, offering new insight into how HFD drives early intestinal maladaptation underlying metabolic dysfunction and increased colorectal cancer risk.

Aliluev A, Tritschler S, Sterr M, Oppenländer L, Hinterdobler J, Greisle T, ... , Theis FJ, Lickert H, Böttcher A. Diet-induced alteration of itestinal stem cell function underlies obesity and prediabetes in micne. Nat Metab. 2021; PMID: 34552271

Beyaz S, Mana MD, Roper J, Kedrin D, Saadatpour A, Hong SJ, Bauer-Rowe KE, Xifaras ME, Akkad A, ..., Yuan GC, Orkin SH, Sabatini DM, Yilmaz ÖH. High-fat diet enhances stemness and tumorigenicity of intestinal progenitors. Nature. 2016; PMID: 26935695;

Abstract No. T39

TET2-mutant clonal hematopoiesis drives inflammation-associated hepatocarcinogenesis in a murine NAFLD model

Sena Alptekin[1, 2, *], *Lina Merg*[1], *Esther Rodríguez-Correa*[1], *Elif Ceren Saf*[1], *Chaofan Fan*[1], *Peng Shen*[1], *Robert Vanner*[3], *Mathias Heikenwälder*[1,4], and *Michael Milsom*[1]

[1] German Cancer Research Center (DKFZ), Heidelberg
[2] Heidelberg University Hospital
[3] Princess Margaret Cancer Centre, Toronto, Canada
[4] The M3 Research Institute, Tübingen

*Presenting author

Clonal hematopoiesis (CH), marked by somatic mutations in hematopoietic stem and progenitor cells (HSPCs), increases with age and is frequently driven by alterations in epigenetic regulators such as TET2. While TET2-mutant CH is associated with a systemic pro-inflammatory phenotype and heightened risk of cardiovascular and hematologic diseases, its role in solid tumor development remains unclear. Non-alcoholic fatty liver disease (NAFLD), a growing global health burden, is a major precursor of hepatocellular carcinoma (HCC). Given the importance of chronic inflammation in NAFLD–HCC progression, we hypothesized that TET2-mutant hematopoietic clones promote liver tumorigenesis.

To test this, we used a tamoxifen-inducible CH model (Scl-CreERT;Tet2^flox/flox), initiating Tet2 loss at the HSPC level. Male mice were fed a high-fat, high-sugar Western diet, and liver tumor development was assessed after 6 months. Tet2-CHIP mice exhibited a markedly higher incidence and severity of HCC compared to wild-type controls, indicating a synergistic interaction between mutant hematopoiesis and liver injury.

In the bone marrow, Tet2-CHIP mice showed expansion of the long-term HSC compartment, consistent with altered stem cell homeostasis. Immune profiling revealed a marked increase in $CD8^+$ T cells in both liver and bone marrow, implicating these cells in the inflammatory response.

To further dissect the contribution of specific hematopoietic lineages to disease progression, we performed variant allele frequency (VAF) analysis across sorted hematopoietic populations in blood, bone marrow, and liver. As expected and fitting with existing literature, myeloid cells exhibited the highest Tet2 VAF. However, surprisingly, myeloid VAF negatively correlated with HCC severity, whereas $CD8^+$ T cell VAF showed a strong positive correlation, pointing to a possible effector role for mutant T cells in driving liver inflammation and carcinogenesis.

These findings identify Tet2-mutant CH as a mechanistic driver of liver cancer and highlight how stem cell–confined mutations can influence distant tissue inflammation and malignant transformation.

Abstract No. T40

Modeling type 2 alveolar epithelial cell biology and disease using patient-derived iPSC

Killian Hurley

Royal College of Surgeons in Ireland, Dublin

abstract not available

Abstract No. T41

Sotatercept as a novel therapy for pulmonary arterial hypertension: Attenuation of fibrosis and contractility in iPSC-derived smooth muscle cells

Aileen Schmidt[1,*], *Leonie Malhofer*[1], *Laurien Czichon*[1], *Gioia Bartsch*[1], *Alexandra Haase*[1], *Astrid Weiß*[2], *Julia Baldauf*[2], *Ralph Schermuly*[2], *Ulrich Martin*[1], and *Ruth Olmer*[1]

[1] Hannover Medical School (MHH)
[2] Justus-Liebig University Giessen

*Presenting author

Heritable pulmonary arterial hypertension (HPAH) is a life-threatening disease characterized by irreversible pre-capillary pulmonary artery remodeling and increased arterial pressure, leading to severe cardiovascular and respiratory symptoms. Sotatercept, a novel FDA-approved therapeutic drug, offers new hope for high-risk patients with HPAH. This drug acts as an activin signaling inhibitor, rebalancing the pro- and anti-proliferation pathways in vascular cells by modulating the TGFβ pathway.

However, the full mechanism of action of Sotatercept, particularly on specific vascular cell types and on different HPAH phenotypes, remains unclear. To address this, we established an HPAH in vitro model using patient-specific induced pluripotent stem cells (iPSCs), which serve as an attractive source for functional vascular smooth muscle cells (iSMCs). We successfully differentiated two HPAH-iPSC lines, each carrying different heterozygous mutations in the TGFβ pathway-related BMPR2, into functional iSMCs. In time-course experiments, we recapitulated the pro-proliferative HPAH phenotype, which was effectively mitigated by Sotatercept as expected. Furthermore, HPAH-iSMCs demonstrated significant contraction in a collagen contraction assay and enhanced ROCK activity upon activinA stimulation, which was alleviated by Sotatercept. Additionally, HPAH-iSMCs produced fibrotic filaments, including collagen I and fibronectin, which were counteracted by Sotatercept. Bulk RNA sequencing and western blot analysis provided molecular evidence for the rescue of the vasoconstrictive and fibrotic phenotype, with SMAD2/3 and non-SMAD signaling playing a dominant role.

These new insights into the mode of action of Sotatercept have significantly advanced our understanding of HPAH pathogenesis and may make add-on medication, such as other vasodilators, expendable.

Abstract No. T42

The lung microvasculature promotes alveolar type 2 cell differentiation via secreted SPARCL1

Paolo Panza[1,2,*], *Hyun-Taek Kim*[2,3], *Till Lautenschläger*[2,4], *Janett Piesker*[2], *Stefan Günther*[2], *Yousef Alayoubi*[2], *Ondine Cleaver*[5], *Mario Looso*[2], and *Didier Stainier*[2]

[1] Justus Liebig University Giessen
[2] Max Planck Institute for Heart and Lung Research, Bad Nauheim
[3] Soonchunhyang Institute of Medi-bio Science, Cheonan, Republic of Korea
[4] Institute for Cardiovascular Regeneration, Goethe University Frankfurt am Main
[5] UT Southwestern Medical Center, Dallas, US

*Presenting author

Lung endothelial cells (ECs) and pericytes are closely juxtaposed with the respiratory epithelium before birth and thus may have instructive roles during development. To test this hypothesis, we screened EC-secreted proteins for their ability to alter cell differentiation in alveolar organoids. We identified secreted protein acidic and rich in cysteine-like protein 1 (SPARCL1) as an extracellular matrix molecule that can promote alveolar type 2 (AT2) cell differentiation *in vitro*. SPARCL1-treated organoids display *lysozyme* upregulation and a doubling in the number of AT2 cells at the expense of intermediate progenitors. SPARCL1 also induces the upregulation of nuclear factor κB (NF-κB) target genes, and suppression of NF-κB activation in lung organoids blocked SPARCL1 effects. NF-κB activation by lipopolysaccharide (LPS) was sufficient to induce AT2 cell differentiation; however, pharmacological inhibition of the pathway alone did not prevent it. These data support a role for SPARCL1 and NF-κB in alveolar cell differentiation and suggest a potential value in targeting this signaling axis to promote alveolar maturation and regeneration.

Abstract No. T43

Differentiation of airway epithelial basal cells into organotypic mucociliary tissues

Janna Nawroth

Helmholtz Munich

Airway epithelial basal cells, whether primary or iPSC-derived, can be readily differentiated into airway epithelial tissues using state-of-art air-liquid-interface (ALI) cultures. However, as these protocols are far from being standardized, different culture media generate different phenotypes, which have been described in terms of their cellular composition and barrier integrity. Missing thus far has been a quantitative and comparative assessment of their mucociliary clearance function, which is key to airway health and, when defective, contributes to major respiratory diseases, including asthma, COPD and cystic fibrosis. Understanding if current differentiation protocols achieve organotypic airway tissues with human-like mucociliary clearance is important for studies of airway disease, regeneration, and aging alike. Here, I present our latest work on addressing this knowledge gap by (1) experimentally identifying and computationally modeling the structure-function relationships that underlie mucociliary clearance in healthy human and rat large airways, and (2) assessing these structure-function relationships in human primary airway ALI cultures. We find that rodent mucociliary clearance is significantly less effective compared to humans, and that most culture media replicate a rodent-like mucociliary clearance phenotype due to a variety of ciliary beat defects that are unique to each medium condition. We show that such analysis can inform protocol choice and development, aid in the comparison of primary versus iPSC-derived tissues, and deliver new insights into airway disease mechanisms.

Abstract No. T44

Aberrant basaloid cells and the fortuitous discovery of a regeneration-associated cell state

Jonas Schupp

MH Hannover

Idiopathic pulmonary fibrosis (IPF) is a fatal interstitial lung disease characterized by progressive scarring driven by maladaptive epithelial repair. Using single-cell profiling, we had previously identified an unrecognized population of aberrant basaloid cells, which coexpress basal, mesenchymal, developmental, and senescence markers and localize to fibrotic lesions. Aberrant basaloid cells have been extensively validated and play a central role in seemingly any fibrotic lung disease. Recently, through ex vivo longitudinal profiling, we demonstrate that these cells emerge from alveolar epithelial cells during injury and share features with transient regeneration-associated states seen under normal repair conditions. Epigenetic profiling reveals hallmarks of AP-1–mediated injury memory, linking persistent activation of this program to fibrosis. These findings uncover a continuum of epithelial plasticity, providing insights into the origins of aberrant basaloid cells and highlighting potential therapeutic targets to restore adaptive regeneration in IPF.

Abstract No. T45

Wnt/Planar cell polarity signaling orchestrates ciliogenesis and cell-cycle exit during endocrine lineage commitment

Tobias Greisle[*], *Anika Böttcher*, *Minas Schwager*, *Michael Sterr*, *Ingo Burtscher*, and *Heiko Lickert*

Helmholtz Center Munich

*Presenting author

In vitro generation of stem cell-derived islets (sc-islets) is a powerful tool to model pancreas development in a human system which is essential for new cell replacement and regenerative approaches in diabetes therapy. However, the critical steps of endocrine progenitor induction, are not completely understood, as it is a highly complex process orchestrated by morphogenetic changes during pancreas development as well as cytoskeletal rearrangements.

One pathway regulating cytoskeletal dynamics and cell fate decisions, is the Wnt/Planar cell polarity (PCP) pathway. Recently, we described the gene Flattop as a reporter and effector of Wnt/PCP signaling. To better understand how morphogenesis and tissue architecture are linked to endocrinogenesis we generated an Insulin (INS-)C-peptide-mCherryxFlattop-2A-H2B-Venus (FVR) double reporter hiPSC line. We performed multiomic analysis spanning epigenetics, transcriptomics and proteomics of FVR+ enriched cells throughout the endocrine induction stage of pancreatic iPSC differentiation. At this stage acquisition of FVR expression coincided with the emergence of endocrine progenitors (EPs) as well as exit from cell cycle. Employing pseudo-time analysis, we resolved endocrine differentiation at enhanced resolution and described novel EP subtypes. In these we identified an EP population expressing a distinct ciliary program driven by the transcription factor *FOXJ1* that involves key components for cilia motility and structure. Upon depletion of *FOXJ1* endocrine cells are still able to form but lack expression of Flattop and other key cilia markers while upregulating processes related to the extracellular matrix. Taken together, our findings identify a crucial time window for cilia restructuring during endocrine induction upon WNT/PCP activation. As cilia are tightly linked to cellular crosstalk and signaling in development, unraveling their specific role upon endocrine induction will further enhance our understanding of the formation of hormone producing cells in vitro and in vivo.

Abstract No. T46

Single-cell multiome uncovers differences in glycogen metabolism underlying species-specific speed of development

Alexandra de la Porte[1,2], *Julia Schröder*[3], *Moritz Thomas*[4], *Max Fernkorn*[2], *Cecile Herbermann*[2], *Johanna Geuder*[5], *Michael Sterr*[6], *Xavier Pastor*[7], *Leslie E. Sanderson*[8], *Tahsin Stefan Barakat*[8], *Wolfgang Enard*[5], *Carsten Marr*[9], *Christian Schröter*[2,3*], and *Micha Drukker*[1,2]

[1] Institute of Stem Cell Research, Helmholtz Munich
[2] Leiden Academic Centre for Drug Research (LACDR), Leiden University, The Netherlands
[3] Max Planck Institute of Molecular Physiology, Dortmund
[4] Institute of AI for Health, Helmholtz Munich
[5] Ludwig-Maximilians-Universität Munich
[6] Helmholtz Diabetes Center, Helmholtz Munich
[7] Core Facility Genomics, Helmholtz Munich
[8] Erasmus MC University Medical Center, Rotterdam, The Netherlands
[9] Institute of AI for Health, Helmholtz Munich

*Presenting author

Embryos from different mammalian species develop at characteristic timescales. These timescales are recapitulated during the differentiation of pluripotent stem cells in vitro. However, specific genes and molecular pathways that modulate cell differentiation speed between mammalian species remain to be determined. We have used single-cell multi-omic analysis of neural differentiation of mouse, cynomolgus and human pluripotent cells to precisely quantify differentiation speed differences between the three species, and to identify regulators for differentiation speed. We demonstrate that species-specific transcriptome dynamics are mirrored at the chromatin level, but that the speed of neural differentiation is insensitive to manipulations of cell growth and cycling. Exploiting the single-cell resolution of our data, we identify glycogen storage levels regulated by UDP-glucose pyrophosphorylase 2 (UGP2) as a species-dependent trait of pluripotent cells, and show that lowered glycogen storage in UGP2 mutant cells is associated with accelerated neural differentiation. The control of energy storage could be a general strategy for the regulation of cell differentiation speed.

Abstract No. T47

NSD1 governs H3K36me2-mediated DNA methylation and drives differentiation of human iPSCs by regulating 'HIDEN' lncRNA expression

Anna Hansknecht[1,2], *Martina Mankarious*[1,2], *Monica Varona Baranda*[1,2], *Esra Dursun Torlak*[1,2], *Wolfgang Wagner*[1,2], *Kira Zeevaert*[1,2], and *Deepika Puri*[1,2,*]

[1] Institute for Stem Cell Biology, University Hospital, RWTH Aachen
[2] Helmholtz Institute of Biomedical Engineering, University Hospital, RWTH Aachen

*Presenting author

Epigenetic regulatory mechanisms, which include histone modifications and DNA methylation, play a central role in development and aging. Dimethylation of H3K36, deposited mainly by the histone methyltransferase NSD1, occurs predominantly in intergenic regions and recruits the DNA methyltransferase DNMT3A to facilitate DNA methylation. Haploinsufficiency of NSD1 results in the overgrowth disorder Sotos syndrome in vivo, which is associated with aberrant DNA methylation signatures and an enhanced epigenetic age. To understand the mechanisms by which NSD1 may regulate development, differentiation and diseases, we generated human iPSC lines deficient in functional NSD1 (NSD1-KO). NSD1-KO cells exhibit a substrate-specific decrease in proliferation, reduced H3K36me2 levels and extensive DNA hypomethylation. Notably, the loss of functional NSD1 altered the differentiation potential of iPSCs, with aberrant endodermal and mesodermal lineage commitment. Further analysis revealed that NSD1 might drive endodermal differentiation by regulating the expression of an endodermal lncRNA 'HIDEN' by mechanisms independent of the regulation of DNA methylation. Our NSD1-KO iPSC lines partially recapitulate the DNA methylation defects associated with NSD1-related disorders (Choufani et al., 2015). Additionally, we uncovered a novel mechanism by which NSD1 may regulate endodermal differentiation of human iPSCs.

Choufani, S., Cytrynbaum, C., Chung, B. et al. NSD1 mutations generate a genome-wide DNA methylation signature. Nat Commun 6, 10207 (2015). https://doi.org/10.1038/ncomms10207

Abstract No. T48

Patient-specific gene correction in LMNA-linked cardiomyopathy using 3D hiPSC-derived heart-tissues and native myocardial slices

Eleonore Baier[1,2,*], *Sofia Ottonello*[1,3], *Elisa Di Pasquale*[4,5], *Christian Kupatt*[1,2], *Gianluigi Condorelli*[4,6], *Julian Grünewald*[1,3], *Christine Maria Poch*[1,2], *Alessandra Moretti*[1,2], and *Karl-Ludwig Laugwitz*[1,2]

[1] Department of Medicine I: Cardiology, TUM Rechts der Isar Hospital, Technical University of Munich
[2] DZHK (German Center of Cardiovascular Research), Munich Heart Alliance
[3] Center for Organoid Systems and Tissue Engineering (COS), Technical University of Munich
[4] Department of Cardiovascular Medicine, Humanitas Research Hospital, Milan, Italy
[5] Institute for Genetic and Biomedical Research, UOS of Milan, National Research Council of Italy
[6] Department of Biomedical Sciences, Humanitas University, Milan, Italy

*Presenting author

Genetic cardiomyopathies can progress to end-stage heart failure, affecting approximately 40 cases per 100,000 individuals, often requiring **heart transplantation**. Currently, therapeutic options to correct or modify disease-causing mutations directly in the human heart do not exist. This fact is partially caused by the lack of human test models to functionally analyze the safety and efficacy of Clustered Regularly Interspaced Short Palindromic Repeats (**CRISPR**)-based **gene editing** approaches ex vivo.

Here, we adopt a translational platform by combining multidisciplinary approaches based on non-viral delivery of next-generation CRIPR editing tools. As an example, we focussed on the correction of two severe dilatative cardiomyopathy (DCM)-causing mutations in the **LMNA** gene. Patient-specific 3D heart tissues derived from human induced pluripotent stem cells (hiPSCs) and native human myocardial slices after transplantation were employed. The primary objective was the precise correction of the pathogenic mutations in the *LMNA* gene using advanced genome editing tools such as **base** and **prime editing**. The bioengineered heart patches reliably reproduced disease-specific phenotypes of mechanical contraction, Ca^{2+} cycling and electrical conduction. Lipid nanoparticles (**LNPs**) were used as a scalable, non-viral vector system to deliver mRNA-based gene correction both **in vitro** and **ex vivo**.

These **clinically relevant model systems** will serve as platforms for the characterization and optimization of CRISPR editor delivery, to study editing efficiencies and precision, as well as their functional impact post-editing. They offer a precise and safe framework for evaluating functional outcomes of therapeutic gene correction for inherited diseases of the heart.

Abstract No. T49

3D transcriptomic reconstruction highlights human heart-forming organoid development as a model of early organogenesis

*Liam Wilson**, *Lika Drakhlis*, *Felix Kleemiß*, *Jana Teske*, *Miriana Dardano*, and *Robert Zweigerdt*

Hannover Medical School (MHH)

*Presenting author

The human embryo undergoes gastrulation and subsequent body plan establishment between 14–21 days post-fertilisation. Difficulty accessing samples has hindered our understanding of this critical window. Despite this, recent in vitro models have made significant progress in replicating the co-development of relevant cell types. However, one of the first morphological processes in human organogenesis—the caudal invagination of the primitive gut tube dorsal to the cardiac mesoderm—remains unexplored. In 2021, we described the human heart-forming organoid (HFO), which recapitulates the co-development of heart, foregut, and vasculature in a robust and reproducible manner. To study the developmental mechanisms from pluripotency to proper heart and foregut patterning at day 10, we performed daily 10x Visium HD spatial RNA-seq on multiple sections, which were spatially contextualised using 3D confocal imaging of whole-mount immunofluorescence-stained and cleared HFOs. This revealed a MIXL1^{+} primitive streak-like population surrounding an ectodermal core following 24 hours of WNT exposure (day 1), leading to a dynamic, gastrulation-like invagination of cells. This population then served as the origin point for endodermal differentiation, with definitive endoderm (FOXA2^{+}/SOX2^{-}) cells migrating inward and gaining SOX2 expression, committing to an anterior foregut (AFE) fate by day 3. From the same region, HNF4A^{+} posterior foregut endoderm (PFE) cells expanded outward by day 4. These events occurred simultaneous to the emergence of endothelial, cardiomyocyte, and cardiac fibroblast lineages. HFOs therefore mimic dynamic gastrulation, followed by migration and development of the foregut and its anlagen in a human system—something previously accessible only in model organisms. We next assessed HFOs' ability to recapitulate in vivo genetically induced phenotypes and confirmed that GATA4^{-}/$^{-}$ HFOs replicated the loss of PFE derivatives observed in GATA4^{-}/$^{-}$ mouse embryos. Together, we provide independent experimental evidence that HFOs model key developmental processes, enabling deeper mechanistic insight into previously inaccessible aspects of human development.

Drakhlis, L., Biswanath, S., Teske, J., Ritzenhoff, K., Franke, A., Manstein, F., Bolesani, E., Kempf, H. and Liebscher, S., 2021. Human heart-forming organoids recapitulate early heart and foregut development. Nature Biotechnology, 39(6), pp.737-746.

Abstract No. T50

Digital reprogramming decodes epigenetic barriers of cell fate changes

Eva Hörmanseder

Institute of Epigenetics and Stem Cells, Helmholtz Munich

Differentiated cells in our body rarely, if ever, change their cell type: They have a cellular memory of their identity which prevents unwanted cell fate changes in healthy organisms which may be circumvented during ageing or in disease. However, vertebrate eggs have the remarkable ability to reprogram cell fates when the nucleus of a somatic cell is transferred to an enucleated egg. The so generated nuclear transfer embryo can erase the cellular memory of the previous cell identity, allowing the establishment of totipotency, and can so give rise to all cell types of the body. In our lab, we use reprogramming via nuclear transfer as a tool to study cellular memory and its contribution to resistance to cell-fate reprogramming. We combine computational approaches with multi-omics and classic embryology assays to shed light on the so far unknown molecular mechanisms that enable, drive or resist the conversion of a differentiated cell identity to totipotency. Recently, we developed digital reprogramming, a computational approach that can predict if genes are susceptible or resistant to reprogramming in frog and human nuclear transfer embryos using only epigenomic and transcriptomic data as input. We have identified chromatin features that underlie genes resistant to reprogramming and that may contribute to the epigenetic barriers that ensure cell-fate stability.

Abstract No. T51

Reinforcement learning for relapse prediction from sparse measurements in exponentially progressing disease

*Arina Shelashen**, *Friedemann Uschner*, and *Ingmar Glauche*

Technical University Dresden

*Presenting author

Many hematological malignancies respond well to standard induction therapy, but some have a marked tendency to relapse despite achieving initial remission. As one prominent example, Acute Myeloid Leukemia (AML) is a genetically diverse disease with different remission behavior and high relapse rates. Experienced clinicians often recognize underlying patterns and adjust strategies to monitor residual disease levels and potentially prevent relapse. Similarly, reinforcement learning (RL) agents improve their decision-making by learning from experience, suggesting RL as a promising tool for supporting clinical decision-making. In this study, we investigate whether RL agents could determine optimal timings for follow-up assessments during disease remission for early relapse detection, while minimizing unnecessary patient interventions.

We used the Gymnasium toolkit to develop an RL framework for relapse prediction. Residual disease dynamics are simulated using a simple exponential growth model. A stochastic Soft Actor-Critic algorithm was implemented to address the problem. Performance was evaluated based on the final reward value considering time-to-relapse detection, overall intervention frequency, and mean absolute error of relapse prediction.

Despite data sparsity, the RL model achieved an average mean absolute error of approximately 15 days in relapse prediction while requiring only two additional patient assessments. We demonstrated that increasing measurement uncertainty decreases prediction accuracy and necessitates more interventions. Furthermore, a shift in the relapse point distribution within the simulated data leads to a corresponding shift in the measurement distributions performed by the agent. Thus, the model exhibits consistent and expected behavior in response to changes in the setup.

Our findings demonstrate that the developed RL model accurately predicts relapse timing even with sparse input data from simulated scenarios. However, further development incorporating clinically realistic scenarios and biological insights is necessary to enhance its real-world applicability.

Abstract No. T52

Uncovering transcriptional drivers of cell identity with generative modeling and high-throughput reprogramming

Mariia Minaeva[1,*], *Dominik Klein*[1], *Ilia Kurochkin*[2], *Carlos-Filipe Pereira*[2], and *Fabian J. Theis*[1]

[1] Institute of Computational Biology, HMGU, Helmholt Munich
[2] Molecular Medicine and Gene Therapy, Science for Life Laboratory, Lund Stem Cell Center, Lund University, Sweden

*Presenting author

Direct reprogramming by transcription factors (TFs) offers a powerful route for generating defined immune cell types for regenerative medicine and immunotherapy. Unlike induced pluripotent stem cell (iPSC)-based approaches, this strategy bypasses intermediate pluripotency and enables rapid access to rare immune subsets. However, the discovery of effective TF combinations has so far relied largely on trial-and-error, with only a few successful examples (e.g., macrophages and conventional dendritic cells type 1 (*Rosa et al. 2018*), cDC1) identified to date.

Here, we introduce a systematic framework that combines a computational generative model (*Klein et al. 2025*) with REPROcode (*Kurochkin et al. 2025 in revisions*), a high-throughput screening platform for multiplexed testing of TF combinations. This integrated strategy allows us to comprehensively explore the combinatorial TF landscape and to disentangle the individual and cooperative roles of TFs in specifying immune cell identity.

Using this approach, we identify novel TF cocktails that efficiently reprogram cells into distinct dendritic cell (DC) types — key players in anti-tumor immunity and promising candidates for cancer immunotherapy. Beyond the generation of DC types, our analyses also reveal TFs that enhance cDC1 identity or drive specialized DC programs, such as mregDCs. Experimental validation confirms that these predicted cocktails give rise to cells with robust cDC1 or mregDC phenotypes.

In summary, our work establishes a scalable and rational strategy for designing TF-based reprogramming cocktails, accelerating the generation of diverse immune cell types for therapeutic applications.

Rosa, Fábio F., Cristiana F. Pires, Ilia Kurochkin, Alexandra G. Ferreira, Andreia M. Gomes, Luís G. Palma, Kritika Shaiv, et al. 2018. "Direct Reprogramming of Fibroblasts into Antigen-Presenting Dendritic Cells." Science Immunology 3 (30): eaau4292.

Klein, Dominik, Jonas Simon Fleck, Daniil Bobrovskiy, Lea Zimmermann, Sören Becker, Alessandro Palma, Leander Dony, et al. 2025. "CellFlow Enables Generative Single-Cell Phenotype Modeling with Flow Matching." bioRxiv.

Abstract No. T53

High resolution spatio-temporal mapping reveals dedifferentiation niches in mammalian cardiac repair

Andy Shing-Fung Chan[1,*], *Joachim Greiner*[2], *Lisa Marschhäuser*[1], *Tomas Brennan*[2], *Ankit Agrawal*[1], *Helene Hemmer*[1], *Katrin Sinning*[1], *Wing Lam Cheung*[1], *Zafar Iqbal*[2], *Thomas Seidel*[2], *Martin Vaeth*[2], *Peter Kohl*[2], *Franziska Schneider-Warme*[2], and *Dominic Grün*[1]

[1] Würzburg Institute of Systems Immunology, Max Planck Research Group
[2] Institute for Experimental Cardiovascular Medicine, University Heart Center, Freiburg

*Presenting author

The adult human heart has minimal regenerative capacity due to the absence of cardiac stem cells and the limited proliferation of cardiomyocytes (CMs), leading to fibrotic scarring and permanent functional loss after myocardial infarction. To understand regenerative limitations, we reconstructed the spatio-temporal dynamics of lesion repair in infarcted adult mouse hearts from day 1 to day 56 post-injury by integrating single-cell RNA sequencing (scRNA-seq) with high resolution-spatial transcriptomics (Xenium). This approach revealed dynamic heterocellular interactions shaping wound healing and scar formation. Ligand-receptor analysis across spatially distinct zones highlighted shifting cell communication niches over time.

Importantly, we uncovered a rare population of dedifferentiating CMs emerging in the myocardium as early as day 1 post-injury, expressing embryonic and cell cycle genes. Signals from endothelial and immune cells in the border zone and endocardium appeared to support this plasticity. We identified innate lymphoid cell type 2 (ILC2) as a key immune niche component promoting CM dedifferentiation through BMP-7 signaling, distinct from hypertrophic responses that dominate typical injury repair. Alongside macrophages and endothelial cells, ILC2s form a "dedifferentiation niche" that reactivates progenitor-like programs in CMs. In vitro, treating postnatal day 7 (P7) CMs with BMP-7 and niche ligands induced cell cycle entry, cytokinesis and progenitor gene expression. Similarly, ex vivo culture of human cardiac tissue slices with these ligands also led to sarcomeric disassembly, proliferation and upregulation of embryonic CM markers, indicating a conserved regenerative mechanism across mouse and human.

Our findings suggest that remnant regenerative pathways persist even in non-regenerative adult animals, despite remain underutilized due to niche cell scarcity. These insights offer new therapeutic avenues toward scarless heart repair by reactivating endogenous CM plasticity.

Abstract No. T54

Napari-organoid-counter: An open-source tool for high-throughput organoid detection in brightfield microscopy

Christina Bukas[1,*], *Fenja See*[1], *Carina Steinchen*[1], *Gowtham Boosarpu*[2], *Sara Asgharpour*[1], *Gerald Burgstaller*[1], *Mareike Lehmann*[1,2], and *Marie Piraud*[1]

[1] Helmholtz Munich
[2] Philipps-University Marburg

*Presenting author

Organoids are increasingly used in stem cell research and regenerative medicine as physiologically relevant 3D models. However, scalable and standardised tools for analysing organoid morphology in microscopy images remain limited. We present the **napari-organoid-counter**, an open-source plugin for the napari image viewer, designed to support fast and reproducible detection and quantification of organoids in brightfield microscopy images.

The plugin integrates deep learning-based detection models and provides an intuitive interface for visualising and annotating organoids across diverse experimental conditions. It was developed as part of the MultiOrg initiative [1], which released the largest curated dataset of murine lung organoids to date. While the current version of the plugin focuses on organoid detection, future development will extend its capabilities to phenotype classification.

The **napari-organoid-counter** is freely available [2,3]. It is already being adopted by labs for high-throughput organoid screening, with more than 360 installs [2]. We invite the stem cell community to explore, contribute to, and help shape its future development.

[1] Bukas, C., Subramanian, H., See, F., Steinchen, C., Ezhov, I., Boosarpu, G., ... & Piraud, M. (2024). MultiOrg: A Multi-rater Organoid-detection Dataset. Advances in Neural Information Processing Systems, 37, 95808-95839.

[2] https://www.napari-hub.org/plugins/napari-organoid-counter;

[3] https://github.com/HelmholtzAI-Consultants-Munich/napari-organoid-counter

Strategic sessions presentations

13th GSCN Conference
German Stem Cell Network
15 – 17 October 2025

Strategic sessions presentations: S1 – S8

S1 Reproducible brain organoids for disease phenotyping and screening
Jakob Metzger

S2 High-content phenotypic profiling reveals subpopulation-specific modulation of human microglia by inflammasome-targeting compounds
Johannes Wilbertz

S3 Bioreactor-based innate immunocompetent hiPSC-derived neurospheroids as preclinical models for advanced therapy development
Catarina Brito

S4 BMP, MEK and WNT inhibition: A novel dSMAD-independent paradigm for rapid generation of hiPSC-derived neurons amenable to regional patterning
Peter Reinhardt

S5 Stay calm and clear: Communication strategies for difficult situations
Nele Haasen

S6 iPSC banks for clinical use: Regulatory and ethical challenges and solutions from the perspective of EU COST Action HAPLOiPS and the Norwegian iPSC Biobank for Clinical Applications (NIBCA)
Joel C. Glover

S7 Title not available
Joanis Mytilineos

S8 Applying haplo-selected hiPSC lines and safety switches: Significant steps towards safer ‚off-the-shelf' iPSC-based ATMPs
Gerald G. Schumann

Abstract No. S1

Reproducible brain organoids for disease phenotyping and screening

Jakob Metzger

Max Delbrück Center, Berlin

Brain organoids are powerful tools for studying human-specific aspects of neural development and for modeling neurodevelopmental disorders. However, their application in phenotyping subtle early disease phenotypes and in high-throughput screening has been limited by challenges in reproducibility and scalability.

To address these limitations, we developed micropatterned neural organoids with high structural and cellular reproducibility, designed to capture early stages of human cortical development and to detect disease-associated deviations.

As a proof of concept, we modeled DYRK1A syndrome, a severe neurodevelopmental disorder. Organoids generated from isogenic cell lines carrying patient-specific heterozygous nonsense mutations in DYRK1A exhibited early defects in progenitor expansion and altered neurogenic trajectories, mirroring cellular phenotypes observed in patients.

Our system is compatible with high-throughput assays and enables statistically robust phenotyping. By enabling the early detection of disease phenotypes and underlying pathways, these organoid models provide a powerful platform for mechanistic studies and therapeutic development in neurodevelopmental disorders.

Abstract No. S2

High-content phenotypic profiling reveals subpopulation-specific modulation of human microglia by inflammasome-targeting compounds

Amélie Weiss[1], *Kayhan Alvandipour*[1], *Michaela Segschneider*[2], *Vanessa Frickel*[2], *Michael Peitz*[2], *Sebastian Kallabis*[3], *Mona Mathews*[2], *Brenda Besemer*[4], *Peter Sommer*[1], *Oliver Brüstle*[2,4], and *Johannes Wilbertz*[1,*]

[1] Ksilink, Strasbourg, France
[2] LIFE & BRAIN GmbH, Bonn
[3] Core Facility Translational Proteomics, Institute of Innate Immunity, University Hospital Bonn
[4] Institute of Reconstructive Neurobiology, University of Bonn Medical Faculty & University Hospital Bonn

*Presenting author

Microglia, the resident immune cells of the central nervous system, exhibit substantial phenotypic plasticity in response to environmental stimuli. However, the heterogeneity of human microglial responses, particularly under inflammasome-activating conditions, remains poorly understood. In this study, we applied high-content phenotypic profiling to human iPSC-derived microglia stimulated with LPS and Nigericin to activate the NLRP3 inflammasome. Using a combination of deep learning-based image analysis with a vision transformer model and manual feature extraction, we characterized microglial phenotypes at single-cell resolution. This approach revealed distinct subpopulations whose composition shifted markedly upon inflammasome activation.

Treatment with the NLRP3 inhibitor MCC950 selectively reversed the phenotypic state of a subset of these subpopulations, particularly those associated with inflammasome activity, suggesting that microglial responses can be modulated in a subpopulation-specific manner. Motivated by these findings, we conducted a broader screen of 212 immunomodulatory small molecules. For the most active compounds, we integrated phenotypic profiling with transcriptomic, proteomic, and secretomic analyses. Key phenotypic features, including ASC speck formation, NF-κB nuclear translocation, and cell morphology, emerged as sensitive and early indicators of compound-induced modulation. During the first hours of inflammasome activation phenotypic profiling captured rapid and functionally relevant cellular changes more effectively than transcriptomic or proteomic readouts.

Our findings demonstrate that inflammasome modulation can induce swift and selective remodeling of microglial phenotypes and secretory profiles, while leaving bulk transcriptomic and proteomic signatures largely unchanged. This work highlights the utility of high-content phenotypic profiling for resolving functional microglial heterogeneity and identifying novel subtype-specific modulators of neuroinflammation.

Abstract No. S3

Bioreactor-based innate immunocompetent hiPSC-derived neurospheroids as preclinical models for advanced therapy development

Catarina Brito

ITQB NOVA & iBET, Oeiras, Portugal

A significant hurdle in ATMP development is the activation of the host immune response in response to the therapeutic cells and gene therapy vectors. Human-relevant experimental assays that depict the innate immune tissue response are required to assess the safety and potency of advanced therapeutics.

To meet this need, we introduce a human innate immunocompetent 3D in vitro model of the CNS and interrogate it to address early tissue response to rAAVs. We explored a neural cell culture methodology pioneered by our team, in which neuron-astrocyte interactions are recapitulated in hiPSC-derived 3D neural cell spheroids (neurospheroids). Neural progenitors derived from human induced pluripotent stem cells (hiPSC) were aggregated and differentiated in perfusion stirred-tank bioreactors (STB) into neurospheroids, which are composed of neurons, astrocytes, and oligodendrocytes. This bioprocess was successfully transferred to a miniaturized STB system (Ambr15), allowing parallelization and higher throughput. Furthermore, we have optimized the integration of microglial cells differentiated from hiPSC (iMGL) by refining culture medium composition and cell ratio.

We will discuss how this downscale human stem cell-based 3D brain cell culture incorporates the innate immune component, recapitulates neuroinflammatory hallmarks, and allows parallelization in the preclinical evaluation of gene therapy viral vectors.

Abstract No. S4

BMP, MEK and WNT inhibition: A novel dSMAD-independent paradigm for rapid generation of hiPSC-derived neurons amenable to regional patterning

Peter Reinhardt

AbbVie Deutschland GmbH & Co KG, Wiesbaden

Induced overexpression of Neurogenin 2 (iNGN2) is a valuable strategy to accelerate the conversion of human induced pluripotent stem cells (hiPSCs) into neurons for modeling of neurodegenerative diseases such as Alzheimer's Disease (AD). However, iNGN2 alone generates neurons of a mixed regional identity and with a strong proportion of peripheral neurons, potentially limiting their relevance for modeling diseases of the CNS. We developed an alternative neural induction strategy, which prepatterns hiPSCs before NGN2 induction to obtain a stronger cortical identity. This strategy involved identifying a novel, rapid, minimal combination of signaling factors for neural induction. By combination of inhibition of BMP signaling and MEK signaling, pathways critical for self-renewal and meso-endodermal differentiation were blocked and an additional inhibition of WNT signaling made this paradigm universally applicable. This novel "BMWi" paradigm within 4-6 days generates a homogeneous culture of rostral neuroectoderm, faster and independent of commonly applied dual SMAD inhibition (dSMADi) and is applicable to a wide range of hiPSC lines. Followed by iNGN2, this induction paradigm quickly leads to homogeneous cultures of excitatory cortical neurons. Furthermore, the cells are responsive to regional patterning cues during the short neural induction pulse. This enables generation of neurons from different regions of both the central and peripheral neuron systems, including midbrain dopaminergic, motoneurons and sensory neurons, improving in vitro models for a range of neurological diseases and neurodegenerative disorders. Cortical neurons generated with this paradigm are suitable for an AD-relevant tau aggregation assay, similar in performance to cortical neurons derived from the dSMADi protocol, highlighting their applicability in disease – relevant *in vitro* models.

Disclaimer: *The author is an employee of AbbVie. The design, study conduct, and financial support for this research were provided by AbbVie. AbbVie participated in the interpretation of data, review, and approval of the publication.*

Abstract No. S5

Stay calm and clear: Communication strategies for difficult situations

Nele Haasen

konteam, Füssen

It can be hard to stay calm and convey your message clearly when emotions run high in difficult situations at work. However, it is possible to develop your communication skills in order to handle these challenging situations more effectively. In this workshop, you will learn and practice some basic techniques to help you overcome future communication challenges.

https://www.konteam.de

Abstract No. S6

iPSC banks for clinical use: Regulatory and ethical challenges and solutions from the perspective of EU COST Action HAPLOiPS and the Norwegian iPSC Biobank for Clinical Applications (NIBCA)

Joel C. Glover

Oslo University Hospital & University of Oslo, Norway

No abstract available

https://haplo-ips.eu

https://www.ous-research.no/home/ncs/National%20Core%20Facility/10094

Abstract No. S7

Title not available

Joanis Mytilineos

German Central Stem Cell Donor Registry (ZKRD), Ulm

Abstract not available

Abstract No. S8

Applying haplo-selected hiPSC lines and safety switches: Significant steps towards safer ‚off-the-shelf' iPSC-based ATMPs

Gerald G. Schumann

Paul-Ehrlich-Institut, Langen

Continuous efforts aim at increasing quality and safety of iPSC-based cell- and gene therapy products: On the one hand, ‚Haplo-selected' hiPSC lines are generated to cover frequent HLA haplotypes in specific populations, allowing a broad range of patients to receive an "off-the-shelf" product that is already HLA-compatible or requires minimal immune suppression; on the other hand, drug-inducible safeguards, termed safety switches, are introduced into the genomes of hiPSC lines and target either residual undifferentiated iPSCs, preventing teratomas, or the entire iPSC-derived cell graft to halt adverse events in cell-based therapies. The presentation will provide an overview of the latest accomplishments regarding both measures to improve the safety profile of iPSC-based therapies.

https://haplo-ips.eu

Company presentations

13th GSCN Conference
German Stem Cell Network
15 - 17 October 2025

Company presentations: C01 – C16

C01 Tackling leachables in stem cell bioprocessing: Ensuring safety and scalability in single-use systems for clinical studies
Philipp Nold, Eppendorf SE

C02 Advance your research with genetically diverse human induced pluripotent stem cell-derived models and a novel mRNA-lipid nanoparticle platform to rapidly generate functional forebrain neurons
Katharina Debowski, STEMCELL Technologies GmbH

C03 Forging the future of regenerative medicine with synthetic peptide growth factors
Robert Brownlee, PeptiGrowth

C04 Approaches for optimizing pluripotent stem cell spheroid expansion in multi-liter bioreactors through the use of constant medium perfusion
David Kuninger, Thermo Fisher Scientific

C05 The world's fastest PCR-based mycoplasma detection assay
Sabine Hahnau-Jurth, Biomerieux Deutschland GmbH

C06 Enabling reliable PSC culture for advanced research applications
Hannah Johannsen, Miltenyi Biotec B.V. & Co. KG

C07 The role of full-length laminins in enhancing in vitro cellular microenvironments
Elias Uhlin, BioLamina

C08 Live-cell 3D cell imaging innovations
Marco Benkisser-Petersen, Sartorius

C09 Recent advancements in gentle single-cell handling: The iotaSciences way
Matthias Hoja, iotaSciences

C10 Alt-R™ CRISPR capabilities to accelerate your next breakthrough
K. Julia Duda, Integrated DNA Technologies (IDT)

C11 Cell therapy at Evotec – end-to-end from process development to clinical manufacturing
Matthias Austen, Evotec

C12 What we've discovered: Opening Pandora's box of genomic mutations using NGS technology
Reda Zenagui, Stem Genomics

C13 Commercial media formulations vs chemically defined home-brewed media for iPSC maintenance
Liz Stewart, Qkine

C14 A global vision for cell and gene therapies
Karin Abitorabi, Minaris Advanced Therapies

C15 Reliable 3D cell models – high-yield cultivation meets precision sorting
Markus Uhrig, OLS OMNI Life Science

C16 Enhancing reproducibility in stem cell and organoid research: Strategies for consistent culture and characterization
Volker Arps, bio-techne

Abstract No. C01

Tackling leachables in stem cell bioprocessing: Ensuring safety and scalability in single-use systems for clinical studies

Philipp Nold

Eppendorf SE, Jülich

BioBLU® single-use stirred-tank bioreactors, with their easy handling and out-of-the-box usability, have become the industry standard, enabling scientists to develop robust processes for batch control and minimize patient risks. Their scalability, from small-scale research to commercial manufacturing, ensures that stem cell therapies can be produced efficiently and safely. Extractables and leachables, chemical compounds that can migrate from bioreactor materials into the culture medium, poses significant risks to cell integrity and therapeutic quality.

This talk explores the advancements in single-use bioreactor technology, focusing on the BioBLU HNQ series, which is optimized for clinical and commercial manufacturing. The HNQ variants are based on the well-established BioBLU vessels, coming with comprehensive documentation packages, supporting compliance with regulatory standards and supporting qualification efforts during audits.

By addressing the leachable challenge and leveraging the benefits of single-use systems, this presentation aims to provide insights into how these innovations are driving breakthroughs in stem cell bioprocessing.

Abstract No. C02

Advance your research with genetically diverse human induced pluripotent stem cell-derived models and a novel mRNA-lipid nanoparticle platform to rapidly generate functional forebrain neurons

Andrew Gaffney[1], *Robert Judson*[1], *Dorah Owango*[1], *Minoo Karimi*[1], *Savitha Deshmuk*[1], *Gary Braun*[1], *Charlotte Ellis*[1], *Erin Knock*[1], *Katharina Debowski*[2,*], *Allen C. Eaves*[1,3], *Sharon A. Louis*[1], and *Arwen L. Hunter*[1]

[1] STEMCELL Technologies Inc., Vancouver, BC, Canada
[2] STEMCELL Technologies Germany GmbH, Cologne
[3] Terry Fox Laboratory, BC Cancer, Vancouver, BC, Canada

*Presenting author

Join us for an insightful session on our latest advancements in cell line generation, featuring four healthy control induced pluripotent stem cell (iPSC) lines which represent a significant step forward in the field, aligning with and advancing beyond the established ISSCR guidelines for stem cell research. By addressing variability and enhancing model relevance, our panel of human iPSC lines not only sets new benchmarks in research quality and ethical standards, but also increases control diversity, ultimately improving the relevance of findings across diverse ethnic and genetic backgrounds.

We also developed iPSCdirect™, a ready-to-use, single-cell cryopreserved format derived from the SCTi003-A healthy control iPSC line. These cells allow to bypass prolonged culture and line establishment, saving both time and resources. iPSCdirect™ cells remove the variability of in-house iPSC banks, offering a reproducible, high-quality source for diverse applications such as disease modeling, drug screening, and toxicity testing.

We also will introduce a novel, integration-free, mRNA-lipid nanoparticle system designed for the efficient and reproducible generation of highly pure neurons from human pluripotent stem cells (hPSCs). Within 6 days, this system enables rapid neuron differentiation with high purity and functional maturation, promotes sustained synaptic activity, simplifies workflows, and reduces common challenges associated with viral methods. The resulting neurons exhibit hallmark electrophysiological and morphological features critical for neurological disease modeling and neuropharmacology.

Our platforms are completed by a growing portfolio of hPSC-derived, ready-to-use differentiated cell types.

Together, these platforms have the ability to help advancing research with high-quality, reproducible, and scalable human models to enhance the fidelity of disease models and increase the efficiency of drug screening pipelines.

Abstract No. C03

Forging the future of regenerative medicine with synthetic peptide growth factors

Robert Brownlee[1,*], and *Kazuki Shigematsu*[2]

[1] Opes Diagnostics, Market Harborough, UK
[2] PeptiGrowth, Tokyo, Japan

*Presenting author

Growth factors derived from conventional sources such as fetal bovine serum (FBS) and recombinant proteins have supported the advancement of regenerative medicine. However, these sources show critical limitations, including variability, animal-derived risks, limited scalability, and high manufacturing costs, which restrict their ability to meet the demands of emerging technologies in the field.

To address these issues, PeptiGrowth Inc. has developed a series of fully chemically synthesized peptide-based growth factors (PG-peptides) that mimic the biological activities and functions of conventional growth factors and cytokines. Free from any animal-derived components, PG-peptides offer comparable capabilities in receptor activation, cell proliferation, and differentiation. Additionally, they provide significant advantages such as improved ease of use, longer shelf life, consistent quality, and reduced costs in R&D, manufacturing, and quality control.

We have currently launched 12 products (e.g., HGF, VEGF, Wnt3a, FGF2 and KGF), and plan to launch 2 more (PDGF-AA and IL-15) by the end of 2025.

We have also generated successful data demonstrating the development of nerve organoids, lung alveolar organoids, pancreatic beta cells, and iPS-derived NKT cells using PG-peptides in the same manner as with conventional growth factors.

We believe that our synthetic peptide growth factors will bring innovative advances to the fields of regenerative medicine and cell therapy. In this industrial presentation, we will present further details on PG-peptides and their applications.

Abstract No. C04

Approaches for optimizing pluripotent stem cell spheroid expansion in multi-liter bioreactors through the use of constant medium perfusion

David Kuninger

Thermo Fisher Scientific, Frederik, US

To meet the needs of clinical manufacturing processes requiring billions of pluripotent stem cells (PSCs) for generation of allogenic cell therapy products, the development of PSC culture methods in large volume bioreactors is critical. We`ve previously shown how PSCs can be expanded as spheroids in 3L stirred-tank reactors (STRs). These types of bioreactors use impellers that continuously mix the medium but also generate shear stress that may negatively affect PSC health. Current protocols relying on gravity sedimentation of PSC spheroids and manual aspiration of spent medium are impractical beyond a 3L culture scale. STRs can be combined with perfusion apparatuses to constantly supply fresh medium and mitigate the need to manually exchange spent medium. Here, we describe the optimization of PSC spheroid growth in liter-scale bioreactor cultures by balancing stir speeds, shear stress effects, and medium exchange rates using constant perfusion of StemScale PSC Suspension Medium. Cells were seeded into STRs at low stir speeds to promote spheroid formation. The RPM was then gradually increased to prevent spheroid aggregation and maintain culture homogeneity. Shear was detrimental to spheroid growth at high speeds, though this could be minimized with shear protectants (e.g., Pluronic). Our results indicate that the addition of 0.1 – 0.2% Pluronic enabled spheroids to grow larger and more uniform in size, resulting in greater cell yields. We evaluated spheroid growth by using tangential flow depth filtration (TFDF) and alternating tangential flow (ATF) perfusion systems for medium exchanges. We determined both systems being able to support spheroid expansion with similar efficiencies. With these optimized parameters we observed yields of up to 25 billion PSCs (12x106 cells/mL, equaling ~80-fold expansion) capable of maintaining pluripotency (>95% OCT4+/NANOG+ cells) after 10 days in culture. These results demonstrate that PSC culture in perfused STRs is an effective means to improve PSC expansion workflows.

Abstract No. C05

The world's fastest PCR-based mycoplasma detection assay

Sabine Hahnau-Jurth

Biomerieux Deutschland GmbH, Nürtingen

Mycoplasmas are small parasitic mollicutes which are known as contaminants of cultured cells. Infected cells show reduced metabolism, inhibition of viability as well as limited reproduction and differentiation. Also, chromosomal aberrations can regularly be observed due to mycoplasma nucleases. In cell and gene therapy, these effects result in a decrease of yield and quality of the therapeutic product and does not only lead to significant financial losses but bears also an essential risk for patients' health. **Therefore, it is mandatory specifically for CGT to have a quick testing system available which provides reliable results for small sample volumes within a short time.**

Biomerieux's **GMP-conform BIOFIRE® Mycoplasma** testing solution is a small, closed pouch system which provides results within **less than 60 minutes**. The test requires only a few minutes hands-on time and can easily be conducted any time in between the normal laboratory routine by anyone without any PCR-experience. It does not even need a specific lab space or safety precautions and detects reliably more than 130 different mycoplasma, acholeplasma and spiroplasma species, including those required by the Pharmacopeias at the requested **sensitivity of 10 CFU/ml**.

All reaction steps, including sample lysis, nucleic acid purification, one multiplex PCR as well as one nested singleplex PCR are successively performed within the same closed pouch and are finally evaluated by a **21 CFR 11-compliant software**. For safety, 3 internal controls are included in the pouch, an external control can be obtained separately. All reagents can be kept at room temperature; no cooling or freezing is required.

Biomerieux provides 4 protocols for different sample volumes as well as IQ, OQ, PQ and primary validation. Also, guidance for method suitability testing may be obtained upon request.

Abstract No. C06

Enabling reliable PSC culture for advanced research applications

Hannah Johannsen

Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach

High quality pluripotent stem cell (PSC) lines are the foundation for all downstream applications, including gene editing, differentiation, and disease modeling. This talk outlines the use of the xeno-free culture medium StemMACS™ PSC-Brew XF, developed to maintain high PSC quality across different clones and over extended passages. PSC lines cultured in this system exhibit stable morphology, sustained proliferation, and consistent expression of pluripotency-associated markers, as well as trilineage differentiation potential and genomic integrity.

The medium maintains key physiological parameters including pH, glucose, glutamine, lactate, and ammonia levels within acceptable ranges, which enables flexible feeding schedules such as weekend-free maintenance without compromising cell quality.

By ensuring consistent cell quality and stability, StemMACS PSC-Brew XF enables reliable performance across a wide range of downstream applications. These include compatibility with CRISPR Cas based gene editing workflows, where it supports efficient recovery and expansion of edited monoclonal cell lines, making it a valuable tool for genetic engineering applications.

The medium is also well suited for workflows that involve complex differentiation protocols, such as the generation of cerebral organoids, dopaminergic neurons, hematopoietic stem cells, and endothelial cells, highlighting its versatility in supporting diverse developmental pathways.

Abstract No. C07

BioLamina

The role of full-length laminins in enhancing in vitro cellular microenvironments

Elias Uhlin, *Noah Fereydouni*, *Evan Lee Graham*, *Chiara Annunziata**, *Sam Hobson*, *Sandra Falci*, *Malin Kele*, and *Therese Kallur*

BioLamina, Sundbyberg, Sweden

*Presenting author

Laminins, a family of 16 distinct ECM proteins, are crucial for tissue formation, maintenance, and homeostasis, with expression tightly regulated in space and time. As key components of basement membranes (BM), intact laminins are vital for healthy tissue function, while mutations in laminin genes result in diseases like Pierson syndrome and Epidermolysis bullosa, affecting organs such as muscles, nerves, and skin. Laminin proteins interact with integrin and non-integrin receptors, such as dystroglycan and syndecan, and bind essential growth factors (GF), modulating GF release. As large trimeric proteins, intact laminins provide structural integrity and bioactivity critical for BM functionality. In contrast, fragmented laminins lack these properties, are not naturally produced in healthy tissues, and fail to support tissue homeostasis. Studies show pluripotent stem cells (PSCs) with LAMA5-KO cannot survive without exogenous laminin 521, supporting the essential role of laminin 521 for PSC survival and expansion.

Our study demonstrates the superiority of full-length recombinant laminin-521 in supporting human PSCs compared to fragmented laminins. Full-length laminin-521 significantly enhances PSC survival, proliferation, and migration, enabling single-cell seeding without the need for ROCK inhibitors (1). Importantly, PSCs cultured on intact laminin-521 exhibit enhanced migratory capacity, achieving complete wound closure in migration assays, whereas fragmented laminins support only 50% closure under the same conditions.

These findings highlight the importance of full-length laminin-521 in recreating natural cellular microenvironments, which is crucial for advancing PSC culture techniques, improving differentiation protocols, and refining disease modeling and gene-editing strategies.

(1) Rodin, S. et al. Clonal culturing of human embryonic stem cells on laminin521/E-cadherin matrix in defined and xeno-free environment. Nat. Commun. 2014

Abstract No. C08

Live-cell 3D cell imaging innovations

Nicola Bevan[1], and *Marco Benkisser-Petersen*[2,*]

[1] Sartorius AG, Royston, UK
[2] Sartorius AG, Göttingen

*Presenting author

The use of stem cells to develop relevant disease models has significantly expanded, necessitating advanced instruments and analytical methods to meet the growing demand for comprehensive data. The dynamic economic and scientific landscape of the past year has highlighted the need for instruments that offer enhanced capabilities combined with an easy and intuitive approach. As expectations rise for streamlined methods that deliver high-quality, reproducible data swiftly, Sartorius remains committed to pioneering innovative solutions that advance the development of superior medicines and research. This presentation will showcase the latest advancements in our bioanalytical instruments, designed to support the future of scientific exploration.

Our recent innovations in live-cell analysis enable real-time, high-throughput imaging of 3D cell models, capturing dynamic cellular changes through techniques such as spinning disk confocal and wide-field fluorescence imaging. The intuitive software facilitates the straightforward acquisition and interpretation of complex data. Supported by aligned reagents, comprehensive protocols, and example application datasets, our instruments seamlessly integrate into existing customer workflows.

Abstract No. C09

Recent advancements in gentle single-cell handling: The iotaSciences way

Matthias Hoja

iotaSciences Ltd., Leipzig

iotaSciences delivers efficient automation solutions for gentle single-cell handling through its proprietary GRID technology. Initially adopted worldwide for applications in iPSC handling and gene editing, recent developments have further highlighted our platforms' versatility across a broader range of single-cell workflows. These include organoid research, single-cell lipidomics, and stable recombinant protein production. Our presentation will showcase case studies and experimental outcomes demonstrating the impact of our technology on diverse research areas.

Abstract No. C10

Alt-R™ CRISPR capabilities to accelerate your next breakthrough

K. Julia Duda

Integrated DNA Technologies (IDT), Munich

Empower your genome editing from design to analysis with IDT's complete CRISPR workflow. The Alt-R™ CRISPR Systems were developed through comprehensive research on each component of the technology. Our optimized Cas9 and Cas12a systems, customizable gRNAs, HDR solutions, and analysis tools are backed by expert support for success at every stage of your research.

Abstract No. C11

Cell therapy at Evotec – end-to-end from process development to clinical manufacturing

Matthias Austen

Evotec SE, Göttingen

Off-the-shelf allogenic approaches based on induced pluripotent stem cells (iPSC) offer significant advantages compared to autologous and donor-derived allogenic therapies, including on-demand availability, unlimited availability of starting material, consistent quality of products, and independence from donor/patient cell availability.

Evotec has built a unique, industry-leading end-to-end platform to develop and manufacture off-the-shelf iPSC-based cell therapeutics, fully integrated under one roof, and covering the full bandwidth from conceptualization, process development, upscaling, formal preclinical tox/safety testing and GMP manufacturing of clinical material.

In addition, we conduct R&D to develop innovative, powerful and flexible therapeutic cell type-based platforms to accelerate the generation of product candidates and pipeline building with our partners, with projects focusing e.g. on iPSC-derived islet cells, γδT-cells, macrophages and retinal cells, for the treatment of diabetes, auto-immunity, cancer or retinal degeneration.

Most of our work uses targeted gene editing to achieve improved functionalization and/or hiding of target cells from allograft rejection, and we have established all required capabilities to perform necessary gene edits efficiently and in a GMP compatible manner, including GMP-compatible and documented clonal isolation and banking. This is complemented by a comprehensive range of genetic integrity assays, which in addition to methods covering general genetic intactness at all levels of genomic organization, also allow to fully characterize gene editing outcomes (e.g. on- vs. off-target edits, loci and number of integrations and payload intactness). In addition, advanced *in vitro* and *in vivo* models with human primary immune cells are available to assess the effectiveness of immune shielding interventions, and by accessing the know-how of disease experts across Evotec, newly generated therapeutic cells are tested using suitable state-of-the art cell-based or animal disease models.

This presentation will provide an overview of our technical capabilities and discuss data from some ongoing RnD projects aimed at the generation of innovative allogenic iPSC-derived therapeutics.

Abstract No. C12

What we've discovered: Opening Pandora's box of genomic mutations using NGS technology

Reda Zenagui

Stem Genomics, Grabels, France

This study analyzed **248 samples** using the **NGS-based StemSeq™ panel** to assess the genetic integrity of induced pluripotent stem cells (iPSCs). Several low-frequency somatic variants were found to significantly increase in variant allele frequency (VAF) over successive passages, suggesting a selective growth advantage and raising safety concerns regarding clonal expansion during iPSC culture. One of our key findings was the detection of variants with **variant allele frequencies (VAFs) as low as 1%**, enabling us to distinguish between somatic and germline mutations.

In some cases, variants were detected at VAFs close to 50%, making it difficult to determine whether they originated from the **donor germline or emerged somatically before or during reprogramming.** This ambiguity complicates variant classification and interpretation, particularly in the context of clinical-grade iPSC applications. Our observations underscore the importance of implementing comprehensive genetic screening at the donor cell stage and post-reprogramming to ensure the genomic integrity of iPSC lines.

As expected, **TP53 mutations** which are well-documented drivers of genomic instability, were identified in our dataset. In contrast, the contribution of **BCOR mutations** is less well understood. While only one previous study has reported on BCOR variants in iPSCs, our analysis revealed several additional, previously unreported mutations. We propose explanations why these mutations were not detected in earlier studies, providing novel insights on the genetic risks posed by BCOR alterations.

Overall, our findings emphasize the urgent need for highly sensitive, broad-spectrum genetic testing throughout the iPSC production workflow. Such approaches are essential to ensure the safety, stability, and functional reliability of iPSCs, particularly for use in gene and cell therapy.

Abstract No. C13

Commercial media formulations vs chemically defined home-brewed media for iPSC maintenance

*Liz Stewart**, and *Leighton Sneade*

Qkine, Cambridge, UK

*Presenting author

Induced pluripotent stem cells (iPSCs) are a powerful tool in regenerative medicine, disease modeling, and drug discovery due to their ability to self-renew indefinitely and differentiate into all cell types of the body. A critical factor in the successful culture and application of iPSCs is the formulation of the media used to maintain their pluripotency and viability.

Commercially available media, such as TeSR E8, Essential 8 (E8), and NutriStem, have become widely adopted due to their optimized performance and standardized composition. These formulations are typically tested across multiple iPSC lines and validated to maintain expression of pluripotency markers, stable karyotypes, and robust differentiation capacity. However, the proprietary compositions and high cost can be limiting for laboratories. While in-house media can substantially reduce culture costs, concerns remain regarding their consistency, scalability, and ability to support long-term pluripotency and reproducibility across cell lines.

In this report, we evaluated various culture media for human pluripotent stem cell (iPSC) maintenance. Using the Axion Omni iPSC were imaged every hour after an initial settling period of 15 minutes.

Our findings show that E8-like media, formulated with Qkine FGF2-G3 (145 aa) and TGF-β1, performs comparably to the xeno-free and chemically defined TeSR™-E8™ and Essential 8 (E8) media. When combined with recombinant human vitronectin, this formulation supports the stable maintenance of iPSCs without affecting their proliferation rate or morphological quality. The ability to maintain iPSCs long-term without compromising safety or functionality in this system can significantly reduce the economic burden of large-scale cell expansion for various downstream applications.

Abstract No. C14

A global vision for cell and gene therapies

Karin Abitorabi

Minaris Advanced Therapies, Ottobrun

*Presenting author

The market growth of Cell and Gene Therapy (CGT) demands experienced Contract Development and Manufacturing Organization (CDMO) partners that have the track record, global footprint, end-to-end services, and platform technologies to enable us to reach more patients.

CDMOs need to deliver, be good long-term partners, and establish and functionalize manufacturing and testing platforms that reduce COGS (cost of goods sold) and vein-to-vein time, thus enabling scalable commercial manufacturing.

Minaris Advanced Therapies was established by combining two businesses acquired in early 2025 by a New York-based investment firm, Altaris Capital. This new company brings together two industry leaders under a unified vision to create the largest and most experienced CGT focused CDMO globally, with 25 years of experience, 11 sites globally, 82 cleanrooms and 4 commercial sites in Japan, Germany and the U.S.A.

This, coupled with extensive Cell and Gene Therapy and Viral Vector development (Oxgene) manufacturing and testing capabilities enable Minaris Advanced Therapies to serve the needs of CGT Therapeutic companies at all stages and all geographies now and into the future. This unified global company will also streamline and derisk tech-transfers between sites through standardization of best-in-class digital and equipment solutions.

We are creating a state-of-the-art global CGT CDMO that truly moves the needle for the industry, answering the call for a global, commercial ready, development and manufacturing partner that works with therapeutic companies to deliver Cell and Gene Therapies to more patients.

Abstract No. C15

Reliable 3D cell models – high-yield cultivation meets precision sorting

Markus Uhrig[1,*], *Amir Keric*[1], *Andreas Friese*[1], and *Allysa Stern*[2]

[1] OLS OMNI Life Science, Bremen
[2] Cell Microsystem Inc., Durham, US

*Presenting author

The conventional 2D cell culture is limited in mimicking tissue and organ complexity. 3D cell models, in contrast, hold great promise as in vitro disease models, imitating the in vivo microenvironment. Despite these advantages, traditional methods for generating spheroids and organoids face scalability issues, are resource inefficient, and require manual manipulation to produce assay plates prone to reproducibility problems. Here we introduce a high-yield workflow for cultivating and sorting cells and 3D aggregates using our *CERO 3D Incubator & Bioreactor* and *CellRaft AIR System*. The CERO 3D is a stand-alone device that supports growth of various cells, including stem cells, spheroids, and organoids, by controlling temperature, pH, CO_2, and minimizing shear stress. This improves viability and maturation, reduces apoptosis, and diminishes spontaneous differentiation of iPSC/ESC while significantly lowering running costs. A rapid alternating clockwise/anticlockwise tube movement with specially designed CEROtubes promotes an upward stream of medium, gently floating cells, spheroids, and organoids. We paired the CERO 3D's spheroid production with CellRaft Technology to enable an automated workflow for generating single spheroid assay plates. Spheroids grown in the CERO 3D were seeded on the CellRaft Array, a custom consumable providing spatial segregation of thousands of spheroids and imaged in brightfield and fluorescence using the CellRaft AIR System. With CellRaft Cytometry software, CellRafts containing single spheroids are sorted by phenotypic and morphologic features, including diameter, morphology, and fluorescence. Once identified, the CellRaft AIR system isolates the desired CellRafts into 96-well plates for downstream use. Together, the CERO 3D and CellRaft Technology provide optimal conditions for large-scale spheroid/organoid production and a reliable, automated solution for generating custom assay plates that ensure consistency across replicates and screening applications.

Abstract No. C16

Enhancing reproducibility in stem cell and organoid research: Strategies for consistent culture and characterization

Volker Arps

bio-techne, Wiesbaden

Reproducibility is a foundational principle in academic research, yet stem cell and organoid studies are frequently challenged by variability in donor sources, culture reagents, and experimental conditions. These inconsistencies can compromise data integrity, hinder cross-study comparisons, and delay translational progress. This presentation outlines practical, evidence-based strategies to improve consistency across the entire stem cell and organoid workflow—from initial cell culture to final phenotypic analysis.

Drawing on the peer-reviewed literature, we explore how academic labs can reduce technical and biological variability by adopting validated, traceable reagents and implementing robust quality control practices. Key topics include the impact of donor variability, reagent lot-to-lot differences, and contamination risks, as well as the importance of monitoring cultures throughout the workflow using appropriate markers and assays.

We highlight Bio-Techne's integrated portfolio of solutions designed to support reproducible research, including GMP-grade media, tested extracellular matrices, and AI-engineered cytokines and growth factors. Additionally, we discuss the role of small molecule cocktails in controlled differentiation and pluripotency maintenance, and showcase advanced analytical platforms—such as RNAscope™, ProteinSimple™, and Pala™—for high-resolution cell characterization.

Through case examples and product data, attendees will gain actionable insights into optimizing stem cell and organoid workflows for greater experimental reliability, publication readiness, and translational potential. This session is particularly relevant for researchers engaged in developmental biology, disease modeling, and regenerative medicine, and aims to empower academic labs to achieve higher confidence in their data and accelerate scientific discovery.

Poster presentations

13th GSCN Conference
German Stem Cell Network
15 - 17 October 2025

Poster session I: P001 – P072

Wednesday, 15 October

P001 – P006: Basic, translational & applied hematopoiesis page 132
P007 – P047: Pluripotency, differentiation & PSC-derived models page 139
P048 – P051: Stem cells in aging & disease (cancer stem cells) page 183
P052 – P072: Tissue-derived stem cells & organoids page 188

Even numbers: 17:30 - 18:30 h
Odd numbers: 18:30 - 19:30 h

Poster session II: P073 – P144

Thursday, 16 October

P073 – P081: Computational stem cell biology & systems biology page 212
P082 – P124: Stem cells in disease modeling & drug development page 222
P125 – P144: Stem cells in regenerative therapies page 269

Even numbers: 16:30 - 17:30 h
Odd numbers: 17:30 - 18:30 h

Poster session I: P001 – P072

P001 – P006: Basic, translational & applied hematopoiesis

P001 On the path of in vitro T cell generation: Deciphering the epigenetic character and differentiation behavior of hiPSC-derived T progenitors
Marcel Finke

P002 A microfluidic stem-cell based bone marrow chip for the safety profiling of new drug candidates in pre-clinical drug development.
Selen Kafadar

P003 Leveraging next generation immune cell sourcing utilizing iPSC-derived hematopoietic organoids
Fawaz Saleh

P004 Investigating the role of NME2 in restricting the self-renewal of HSCs to a metabolic niche
Heidrun Schaller

P005 Feeder-free expansion of iPSC-derived NK cells to clinical scale
Daniel Terheyden-Keighley

P006 Generation of human iPSC-derived hematopoietic progenitor cells and differentiation to functional ink cells in a flexible and feeder-free culture system
Tim Wessel

Abstract No. P001

On the path of in vitro T cell generation: Deciphering the epigenetic character and differentiation behavior of hiPSC-derived T progenitors

Marcel Finke[1,*], *Christopher Kressler*[1, 2], *Frederik Hamm*[1], *Ramonique Lim*[1], *Anne Schulze*[1], *Harald Stachelscheid*[3], and *Julia K. Polansky*[1, 2]

[1] BIH Center for Regenerative Therapies (BCRT) at Charité – Universitätsmedizin Berlin
[2] German Rheumatology Research Center (DRFZ), Berlin
[3] BIH Core Unit pluripotent Stem Cells and Organoids (CUSCO) at Charité - Universitätsmedizin Berlin

*Presenting author

T cells are key contributors to the adoptive immune system and in vitro expanded specialized T cell subsets are used in emerging immunotherapy approaches to specifically target a variety of diseases from tumors to autoimmunity. However, the necessary massive expansion of isolated T cells is a major limitation and can lead to a proliferation-acquired senescent cell state and functional instability. Thus, the directed *de novo* generation of specific T cell products *in vitro* from hiPSCs has been proposed as a promising alternative but still faces various critical hurdles.

To advance hiPSC-derived T cell generation for clinical applications, we generated several new hiPSC lines from distinct T cell subsets and established a differentiation protocol that phenotypically mimics thymic T cell development up to the CD4+CD8+ double positive (DP) T progenitor stage via an hematopoietic stem cell (HSC) intermediate step. To confirm successful differentiation of the cells, we analyzed the epigenome of several sorted subsets and intermediary populations from the differentiation assay and compared these profiles to the ones of *ex vivo* isolated counterparts (i.e. cord blood HSCs and thymocyte subsets). Albeit showing a promising combination of surface markers that mimic their thymic counterparts, the hiPSC-derived cells possessed a distinct epigenetic signature. Major differences could already be observed at the HSC stage, which appear to determine downstream differentiation success early on, thus serving as a potential indicator for hiPSC-HSC lymphoid differentiation potential.

Taken together, we generated a large epigenetic dataset, which allowed to assess the quality of *in vitro* generated hiPSC-derived T cells and revealed so far underestimated limitations of commonly-used *in vitro* T cell differentiation systems. These insights will not only improve our understanding of T cell generation in humans, but will also be instrumental to progress towards the hiPSC-derived manufacturing of clinical T cell products for adoptive cellular therapies.

Abstract No. P002

A microfluidic stem-cell based bone marrow chip for the safety profiling of new drug candidates in pre-clinical drug development.

Selen Kafadar[1,*], *Leopold Koenig*[1], *Thi Phuong Tao*[1], *Laurent Juglair*[2], *Susanne Fischer*[2], *Desirée Schubert*[2], and *Annika Winter*[1]

[1] TissUse GmbH, Berlin
[2] F. Hoffmann-La Roche Ltd, Basel, Switzerland

*Presenting author

Hematologic toxicities are frequent dose-limiting side effects encountered during drug development. The use of animal models for assessing bone marrow toxicity is often limited due to the phylogenetic distance between species, which leads to differences in hematopoietic processes and immune system function. To address this challenge, a human bone marrow microphysiological system (MPS) was developed and evaluated for its potential to predict hematopoietic toxicities.

A zirconium oxide scaffold, designed to replicate the structure and surface of trabecular bone, was seeded with bone marrow-derived mesenchymal stromal cells. Human CD34+ hematopoietic stem and progenitor cells (HSPCs) were then introduced into the stromal cell-coated scaffold. The assembled construct was cultured within MPS for up to 31 days. Differentiation and maturation of HSPCs into myeloid, erythroid, and lymphoid lineages were driven by the addition of specific hematopoietic growth factors. Treatment effects were monitored weekly using flow cytometry, erythroid enucleation assays, and cytokine profiling of the culture medium.

Consistent with published data, the presence of pro-inflammatory cytokines, such as IL-1b, altered lineage distribution and cell output. Additionally, the system successfully reproduced the inhibition of erythroblast differentiation caused by bispecific antibodies targeting the transferrin receptor (TfR), a finding previously observed in cynomolgus monkey studies. Results from the chip suggest that antibody-dependent cell-mediated cytotoxicity (ADCC) may contribute to the suppression of erythropoiesis. Exposure to T-cell bispecific antibodies (TCBs) triggered T-cell activation and target cell killing. Notably, the immune response was markedly stronger in an allogeneic versus an autologous setup, indicating that the therapeutic index of TCBs may be underestimated when assessed in allogeneic systems.

Abstract No. P003

Leveraging next generation immune cell sourcing utilizing iPSC-derived hematopoietic organoids

Fawaz Saleh[1,*], *Greta Meyer*[1], *Lucas Rebuffet*[2], *Svenja Gaedcke*[1], *Emilio Valdivia*[1], *Adan Chari Jirmo*[1], *Sabine Buyny*[1], *Maria Michela Santamorena*[1], *Roland Jacobs*[1], *Christine Falk*[1], *Eric Vivier*[2], *Britta Eiz-Vesper*[1], *Robert Zweigerdt*[1], and *Nico Lachmann*[1]

[1] Hannover Medical School
[2] INSERM, Marseille, France

*Presenting author

The number of clinical trials employing hematopoietic stem cells and immune derivatives for cell-based immunotherapies is rapidly increasing. However, the future of these therapies hinges on overcoming critical challenges including donor dependence, immune cell heterogeneity, and the lack of scalable production. Induced pluripotent stem cells (iPSCs) represent a transformative solution providing a limitless and a consistent cell source of immune cells. This work introduces a novel, scalable platform that harnesses iPSC-derived hematopoietic organoids termed Hemanoids, to produce innate immune cells including natural killer cells (NKs), dendritic cells (DCs), and macrophages (MΦ) using simple cytokine cues in 50 ml bioreactors. For NK cell differentiation, Hemanoid were stimulated with IL3, SCF, IL7, FLT3L, and IL15, generating cells expressing NKp46 within distinct CD56+ and CD56- subsets. Transcriptomic analysis revealed a transition from ZEB2 to ZEB1 expression, identifying a developmental trajectory from CD56- to CD56+ NK intermediates. Upon expansion, NKs exhibited enhanced cytotoxicity against K562 target cells and gene expression profiles resembling memory-like NKs. DC differentiation was achieved using GM-CSF, FLT3L, and SR1, yielding populations capable of efficient antigen uptake, processing, and presentation. Functional validation using autologous CMV-exposed patient cells confirmed pro-inflammatory cytokine secretion and memory T cell activation, demonstrating potent functionality. MΦ differentiation was driven by IL3 and M-CSF, yielding 2-3x107 cells/harvest in bioreactors. These MΦ demonstrated efficient phagocytosis of bacterial bioparticles and robust reactive oxygen species production upon stimulation. Functional profiling revealed polarization into both pro- and anti-inflammatory states, resulting in the secretion of CXCL10, CCL17, and IL-10, respectively. In conclusion the results represent a proof-of-concept for advanced manufacturing approaches to source potent immune cells for off-the-shelf immune cell therapies.

Abstract No. P004

Investigating the role of NME2 in restricting the self-renewal of HSCs to a metabolic niche

*Heidrun Schaller**, *Anastasiya Pak*, *Sigrid Uxa*, and *Michael Cross*

Universitätsklinikum Leipzig

*Presenting author

The nucleoside diphosphate kinase “non-metastatic protein 2” (NME2) has been implicated in oncogenic transformation and disease progression in a wide range of cancers, including the myeloid leukemias. NME2 protein accumulates to very high levels in chronic myeloid leukemia cells in which the BCR/ABL fusion protein is active, while the level of Nme2 mRNA in acute myeloid leukemia is variable and associated with prognosis. Interestingly, the prognostic relevance of Nme2 mRNA in AML appears to be particularly significant under conditions of metabolic stress.

Our previous studies using the FDCP-Mix model of multipotent murine stem/progenitor cells confirmed *Nme2* to be a stem-cell specific gene, found that expression is increased under low nutrient conditions, and showed that mNme2 is required for the maintenance of IL-3-mediated JAK-STAT signaling under metabolic stress. Based on these findings, we hypothesize that mNme2 acts as a link between metabolism and signaling in the stem cell niche, tying self-renewal to a metabolically limiting environment, while aberrant expression in myeloid leukemias permits a degree of self-renewal outside these constraints. Specifically, we propose that mNme2 interacts directly with kinases required for self-renewal signaling and then draws on the entire NTP pool to regenerate ATP locally, thus maintaining activity of a select number of kinases under energy-limiting conditions.

We intend to test this hypothesis by identifying binding partners of a tagged mNme2 protein expressed in self-renewing versus differentiating FDCP-Mix cells under various metabolic conditions. An initial comparison of available expression vectors revealed high levels of transgene expression that are likely to lead to promiscuous interaction of the inherently sticky mNme2 protein. To obtain the moderate levels of transgene expression appropriate for identifying specific interactions in self-renewing versus differentiating cells, we have constructed lentiviral vectors containing minimal promoter sequences from the stem-cell-specific Serpina3g gene and the myeloid-specific myeloperoxidase gene.

Abstract No. P005

Feeder-free expansion of iPSC-derived NK cells to clinical scale

*Daniel Terheyden-Keighley**, and *Boris Greber*

Catalent Düsseldorf GmbH

*Presenting author

Natural killer (NK) cells play a critical role in immune surveillance, targeting and eliminating transformed cells before they develop into life-threatening cancers. Unlike T cells, NK cells can be used as allogeneic therapies without the risk of graft-versus-host disease. This enables large-scale manufacturing from a single donor for off-the-shelf therapies. Traditional donor-derived NK cell manufacturing faces challenges such as donor-to-donor variability and random gene insertion, leading to inconsistent production. iPSC-derived NK cells offer a consistent, practically unlimited source material with the advantage of precise, one-time gene-editing. However, current methods using iPSCs often depend on embryoid body-based differentiation and cancerous feeder cells, creating additional complexities for clinical scalability.

An iPSC-derived NK cell platform was developed to address these challenges by enabling feeder-free expansion at clinical scale. This process uses defined media to expand and bank NK precursor cells at small scale before transitioning to cost-effective media for large-scale expansion. The approach eliminates the need for feeder cells and uses GMP-compatible protocols. NK cell purity and functionality were assessed through the expression of NK activation markers and cytotoxic efficacy against K562 cells.

The developed protocol successfully expanded up to several billion NK cells per cm^2 of seeded iPSCs in a feeder-free process. The resulting NK cells exhibited high purity, robust activation marker expression, and potent cytotoxic activity against target cells. To our knowledge, this is the first method to achieve the expansion of iPSC-derived NK cells without feeder cells, using a GMP-compatible process, and at scales sufficient for clinical applications.

This feeder-free iPSC-derived NK cell platform offers a consistent, scalable, and GMP-compatible solution to produce NK cells for clinical use. By overcoming the limitations of traditional donor-derived methods and current iPSC-based approaches, the platform supports the development of off-the-shelf NK cell therapies with enhanced safety, reliability, and manufacturing efficiency.

Abstract No. P006

Generation of human iPSC-derived hematopoietic progenitor cells and differentiation to functional ink cells in a flexible and feeder-free culture system

Abigail Harris Becker[1], *Marcus Bunn*[1], *Lindsay Bailey Steinitz*[2], *Pratima Rayavarapu*[1], *Jeremy De Leon*[1], *Chris Yankaskas*[1], *Vivek Chandra*[2], *Mark Kennedy*[1], *Tim Wessel*[3,*], and *Navjot Kaur*[1]

[1] Thermo Fisher Scientific, Frederick, US
[2] Thermo Fisher Scientific, Carlsbad, US
[3] Thermo Fisher Scientific, Darmstadt

*Presenting author

To meet the needs of clinical manufacturing processes requiring billions of pluripotent stem cells (PSCs) for generation of allogenic cell therapy products, the development of PSC culture methods in large volume bioreactors is critical. We've previously shown how PSCs can be expanded as spheroids in 3L stirred-tank reactors (STRs). These types of bioreactors use impellers that continuously mix the medium but also generate shear stress that may negatively affect PSC health. Current protocols relying on gravity sedimentation of PSC spheroids and manual aspiration of spent medium are impractical beyond 3L culture scale. STRs can be combined with perfusion apparatuses to constantly supply fresh medium and mitigate the need to manually exchange spent medium. Here, we describe the optimization of PSC spheroid growth in liter-scale bioreactor cultures by balancing stir speeds, shear stress effects, and medium exchange rates using constant perfusion of StemScale PSC Suspension Medium. Cells were seeded into STRs at low stir speeds to promote spheroid formation. The RPM was then gradually increased to prevent spheroid aggregation and maintain culture homogeneity. Shear was detrimental to spheroid growth at high speeds, though this could be minimized with shear protectants (e.g., Pluronic). Our results indicate that the addition of 0.1 – 0.2% Pluronic enabled spheroids to grow larger and more uniform in size, resulting in greater cell yields. We further evaluated spheroid growth by using tangential flow depth filtration (TFDF) and alternating tangential flow (ATF) perfusion systems for medium exchanges. We determined both systems being able to support spheroid expansion with similar efficiencies. With these optimized parameters we observed yields of up to 25 billion PSCs (12x106 cells/mL, equaling ~80-fold expansion) capable of maintaining pluripotency (>95% OCT4+/NANOG+ cells) after 10 days in culture. These results demonstrate that PSC culture in perfused STRs is an effective means to improve PSC expansion workflows.

P007 – P047: Pluripotency, differentiation & PSC-derived models

P007 Generation of two induced pluripotent cell lines from patients with an atypical familial form of lung fibrosis
Reem Al Hijailan

P008 A defined monolayer culture system for PSC-derived bRG cells facilitates mechanistic insights into human neurodevelopment
Annasara Artioli

P009 Maximizing iPSC insights: AI-driven live-cell analysis
Marco Benkisser-Petersen

P010 A synthetic, xeno-free surface coating for the culture and differentiation of pluripotent stem cells
Constantin Berger

P011 Non-cryogenic preservation strategies for human iPSC-derived cardiomyocytes
Pranjali Milind Bhagat

P012 Perlecan-conjugated laminin fragment as a next-generation culture substrate for human pluripotent stem cells
Philipp Boder

P013 Directed differentiation of pig pluripotent stem cells to definitive endoderm
Ekin Gökce Cicek

P014 Exploring the impact of telomere-related mutations on iPSC reprogramming in a model of short telomere related pulmonary fibrosis
Amy Duffy

P015 Serum-free differentiation and cultivation of human PSCs into endothelial cells using a closed semiautomated cultivation platform
Kathrin Godthardt

P016 Efficient and scalable 2D based differentiation of human PSCs into hematopoietic stem cells (HSCs) using a xeno- and serum-free cell culture medium
Kathrin Godthardt

P017 Hepatocyte-like cells derived from alpha-1-antitrypsin deficiency-patient iPSCs reproduce disease-associated pathway dysregulation and react to potential drugs
Nina Graffmann

P018 Inter- and intra-donor variability of iPSC generated from human skin biopsies via an accelerated and validated pipeline using non-integrative RNA-based reprogramming
Diana Haberhausen

P019 Advanced hPSC bioprocessing via 3D inoculation from cryopreserved cell stocks
Carlos Alberto Hernandez Bautista

P020 AI-based live imaging of a scaled-up production of human induced pluripotent stem cell-derived cardiomyocytes for in vitro test systems
Sabrina Herrmann

P021 Bioprinted 3D pancreatic ductal tubes: A physiologically relevant model for studying ductal biology, disease mechanisms, and regenerative medicine
Meike Hohwieler

P022 Investigating the role of externally applied cyclic shear stress in early differentiation within iPSC-derived embryoid bodies
Syeda Inaas

P023 Serum- and xeno-free fibroblasts derived from human iPS cells
Yajuan Jiang

P024 Comparing single-cell and clump passaging in iPSC cultures: Implications for genomic stability and standardization
Qianren Jin

P025 Gene editing in hPSCs: The importance of post-editing quality controls
Pia Johansson

P026 Reducing variability in primate iPSCs to improve cross-species comparisons
Antonia Keßler

P027 Differentiation and characterization of astrocyte and endothelial cells from induced pluripotent stem cells of Alzheimer's disease patients which show altered BBB integrity
Sohana Islam Khan

P028 Application of single cell RNA sequencing to characterize transient cell populations and molecular signalling during respiratory differentiation of human pluripotent stem cells
Anna Kuleshova

P029 Novel isthmic-like neural progenitors with high capacity for mid-hindbrain boundary neuron generation in vitro and in vivo
Sarah Lang

P030 Modelling the molecular basis of human speech evolution
Manqi Liang

P031 From research to clinical manufacturing: the reliability of bFGF in supporting iPSCs growth, pluripotency and differentiation
Christine Lansche

P032 Cryopolymer hydrogels as scaffolds for engineered heart tissues in vitro
Ziwei Lu

P033 The landscape of membrane-less organelles in early human differentiation
Gizem Mansour

P034 GMP-compliant iPS cell lines show efficient cardiac induction in a new differentiation platform featuring scalability and enhanced robustness
Soraia Martins

P035 Research on cardiomyocyte maturation through mechanical variation of cell environment
Beate-Sophie Nenninger

P036 UV-micropatterning for high-throughput generation of embryoid bodies
Vera Nischalke

P037 Deciphering the regulatory role of long non-coding RNA in pancreatic development and diabetes
Francesca Piazzalonga

P038 The role of proteolytic and mechanical stress in genome-edited hiPSC-derived cardiomyocytes as a model of myofibrillar myopathy 6
Isabelle Risse

P039 iPSC-derived cardiomyocytes to investigate the role of OCaR2 in abnormal calcium signalling in cardiac arrhythmias
Kristin Rädecke

P040 Synthetic peptide growth factors functionally equivalent to FGF2 and KGF
Kazuki Shigematsu

P041 Comparative analysis of different TBK1-ALS mutations in patient-derived motor neurons to develop a novel screening platform
Jahnavi Srinidhi

P042 Identification of general and tissue-specific mechanisms of Fabry disease using a patient-specific induced pluripotent stem cells model
Ferdinand Teichert

P043 Salt-inducible kinases transduce mechanical forces into the specification of the pancreatic endocrine lineage
Chenglei Tian

P044 Pancreatic alpha and beta cell fate choice is directed by apical-basal polarity dynamics
Chenglei Tian

P045 HERV-H activation disrupts endoderm formation via ZEB2-mediated inhibition of TGF-β/SMAD signaling
Michelle Vincendeau

P046 Rapid generation of oligodendrocyte progenitor cells from neural progenitor cells through 3D spheroid culture and small molecule modulation
Nils Wörner

P047 Production of CAS9 expressing hiPSCs for genetic screen
Ferhat Ali Yaman

Abstract No. P007

Generation of two induced pluripotent cell lines from patients with an atypical familial form of lung fibrosis

Reem Al Hijailan

King Faisal specialist hospital and research center & Alfaisal University, Riyadh, Saudi Arabia

Background: Pulmonary fibrosis is a major disease that leads to progressive loss of lung function, affecting approximately three million patients worldwide with a poor prognosis. Skin fibroblasts isolated from patients offer valuable insights into understanding disease mechanisms, identifying genetic causes, and developing personalized therapies. However, the use of skin fibroblasts to study a disease that exclusively impacts the lungs is often questioned, particularly since lung fibrosis primarily affects the alveolar epithelium. Method: we report the reprogramming of skin fibroblasts from patients with an atypical early-onset form of lung fibrosis into induced pluripotent stem cells (iPSCs), and subsequently into alveolar epithelial cells. This was achieved using a non-integrating approach. Results: show that the reprogrammed cells carry mutations in the calcium-binding protein genes S100A3 and S100A13, leading to diminished protein expression, mimicking patient's cells. Additionally, we demonstrate that the generated patient iPSCs exhibit aberrant calcium and mitochondrial functions. Conclusion: Due to the lack of a suitable animal model that accurately resembles the human disease, generating patient lung cells from these iPSCs can serve as a valuable "disease in a dish" model for understanding the pathogenesis of lung fibrosis.

An atypical pulmonary fibrosis is associated with co-inheritance of mutations in the calcium binding protein genes S100A3 and S100A13. Eur Respir J. 2019 Jul 18;54(1):1802041.

Abstract No. P008

A defined monolayer culture system for PSC-derived bRG cells facilitates mechanistic insights into human neurodevelopment

Annasara Artioli[1,2,*], *Andrea Rossetti*[1,2], *Matteo Gasparotto*[1,2], *Yannick Hass*[1,2], *Anne Hoffrichter*[1,2], *Ryszard Wimmer*[3,4], *Fabio Marsoner*[1,2], *Raquel Perez Fernandez*[1,2], *Alexandre D. Baffet*[3,5], *Philipp Koch*[1,2], *Ammar Jabali*[1,2], and *Julia Ladewig*[1,2]

[1] Central Institute for Mental Health, Mannheim
[2] Heidelberg University/Medical Faculty Mannheim
[3] Institut Curie, Paris, France
[4] CNRS UMR144, Paris, France
[5] Institut national de la santé et de la recherche médicale, Paris, France

*Presenting author

The evolutionary development of the human brain's structural and functional complexity distinguishes Homo sapiens from other species and presents significant investigative challenges. The expansion of basal radial glial (bRG) cells, a primate-enriched progenitor population, is hypothesized to be a key contributor to human brain enlargement and complexity. Previous understanding of bRG cells relied primarily on ex vivo human fetal cortex studies, which were constrained by limited tissue availability. While bRG cells have been identified in human pluripotent stem cell (PSC)-derived brain organoids, their detailed characterization remains challenging in these complex 3D systems. Here, we report the development of defined culture conditions for generating and expanding PSC-derived bRG cells in homogeneous 2D cultures. We established a small molecule cocktail that recapitulates key aspects of the in vivo neural stem cell niche. Immunocytochemistry and transcriptional profiling revealed expression of established bRG-specific markers, and comparative transcriptional analyses demonstrated substantial overlap between our 2D-derived bRG cells and their primary and organoid-derived counterparts. The generated bRG cells exhibited characteristic mitotic somal translocation (MST), a defining behavioral feature of this cell type. Molecular dissection of MST revealed critical role for PAK2 signaling pathway. Furthermore, these in vitro generated bRG cells demonstrated multipotent differentiation capacity, giving rise to diverse cortical layer interneurons and astroglia. In co-culture experiments with forebrain organoids, the bRG cells selectively colonized subventricular zone-like regions and maintained their proliferative and differentiation potential. This 2D protocol provides a simplified experimental platform for investigating bRG cell dynamics and human brain development, offering advantages in accessibility and experimental manipulation compared to existing models.

Abstract No. P009

Maximizing iPSC insights: AI-driven live-cell analysis

Daryl Cole, *Jasmine Trigg*, *Natasha Lewis*, and *Nicola Bevan*

Sartorius AG, Royston, UK

Presenting author – see below

Researchers are keen to derive extensive insights from valuable cells such as induced pluripotent stem cells (iPSCs), while maintaining user-friendly approaches. The Incucyte® Live-Cell Analysis System provides a robust set of computational tools for label-free live-cell analysis, utilizing high-definition phase-contrast imaging. These tools were developed using AI neural network models trained on annotated datasets from various cell types. This data will illustrate the application of these tools in advancing iPSC research.

To produce high-quality in vitro iPSC models, precise concentrations of growth factors are needed to maintain pluripotency, along with plate coatings to support growth. Utilizing Incucyte® AI confluency analysis, various concentrations of FGF-2, a growth factor crucial for optimal growth, were examined. The effects on growth and colony sizes were quantified showing optimal growth at 100 ng/mL whilst ranges 3.13-25 ng/mL caused smaller slower growing colonies. Further data generated with a thermostable variant, FGF2-G3, indicated that labor-intensive daily feeding could be eliminated without compromising cell health. Similar investigations were conducted to explore alternative Xeno-free hydrogel plate coatings.

By leveraging Incucyte® AI Cell Health (AI CH) and Advanced Label Free Classification (ALFC) analyses, deeper insights into cellular responses can be quantified. AI CH offers superior cell segmentation for analysis and when combined with ALFC allows the quantification of morphologically distinct subpopulations. Various pro-inflammatory stimuli were added to iPSC-derived microglia and the resulting growth and morphological characteristics were assessed. Treatment with Interferon gamma (100 ng/mL) caused a 3.5 fold increase in a stellate morphology and increase in phase object area compared to vehicle (1760 ± 45 vs. 1230 ± 40 µm2, respectively at 24 hours), indicative of microglia activation.

The integration of AI techniques for image analysis with the Incucyte® system allows users to extract maximum information from iPSC derived cell models while preserving image analysis workflow simplicity.

This poster will be presented by Marco Benkisser-Petersen.

Abstract No. P010

A synthetic, xeno-free surface coating for the culture and differentiation of pluripotent stem cells

Constantin Berger[1,*], *Stefan Hauser*[2,3], *Simon Widmaier*[4], and *Daniela Zdzieblo*[1]

[1] Fraunhofer Institute for Silicate Research ISC, Translational Center Regenerative Therapies TLC-RT, Würzburg
[2] German Center for Neurodegenerative Diseases (DZNE), Tübingen
[3] Hertie Institute for Clinical Brain Research, University of Tübingen
[4] faCellitate GmbH, Mannheim

*Presenting author

Proper surface coating is essential for maintaining pluripotent stem cells (PSCs) in vitro. Despite significant batch-to-batch variability, undefined composition, and ethical concerns, animal-derived extracellular matrices (ECMs) remain widely used and are still considered the gold standard for PSC culture. Efforts to develop defined, synthetic ECM substitutes have largely failed to date, primarily due to their inability to recapitulate the essential biological functions required for PSC self-renewal and stability.

In this study, we evaluated BIOINSTRUCT™, a synthetic ECM substitute, as a surface coating for the culture of human induced pluripotent stem cells (hiPSCs). IMR90-4 hiPSCs were maintained on BIOINSTRUCT™ or Matrigel™ for ten passages, followed by comprehensive characterization of (1) stem cell properties via gene expression analysis, immunofluorescence staining, flow cytometry, and karyotyping, and (2) pluripotency through spontaneous in vitro differentiation and Germ Layer Score assessment. Additionally, we examined the compatibility of BIOINSTRUCT™ with established protocols for directed differentiation of hiPSCs.

hiPSCs cultured on BIOINSTRUCT™ retained characteristic stem cell morphology, stable proliferation rates, and expression of core pluripotency markers at both the transcriptional (*OCT4*, *SOX2*, *NANOG*) and protein (NANOG, TRA-1-81) levels, alongside a normal karyotype, comparable to cells maintained on Matrigel™. After 10 passages, cells cultured on either substrate successfully differentiated into derivatives of all three germ layers, confirming preserved pluripotency. Notably, Germ Layer Score analysis revealed an ectodermal bias in cultures maintained or differentiated on Matrigel™, which was reversed when using BIOINSTRUCT™. Furthermore, directed neuronal differentiation of hiPSCs on BIOINSTRUCT™ yielded functional cortical neurons, as confirmed by immunocytochemical detection of TUJ and CTIP2.

In summary, our findings demonstrate that BIOINSTRUCT™ supports both the maintenance and differentiation of hiPSCs, highlighting its potential as a xeno-free alternative to animal-derived ECMs for pluripotent stem cell culture.

Abstract No. P011

Non-cryogenic preservation strategies for human iPSC-derived cardiomyocytes

Pranjali Milind Bhagat[*], *Sarkawt Hamad*, and *Kurt Pfannkuche*

Marga-and-Walter-Boll-Laboratory for Cardiac Tissue Engineering, University of Cologne, Cologne

*Presenting author

Human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs) have a wide range of applications from research to clinical trials and more. While a lot of progress has been made in establishing protocols for differentiating of hiPSCs to cardiomyocytes, not a lot has been given to their storage without cryopreservation. One way through which we hypothetically can prolong their viability at 4°C, is by targeting the PDK4-PPARα pathway. It is seen that upregulation of PDK-4 in hibernating mammals preserves their heart during hibernation, by shifting the cells from a glycolytic to a lipid-based metabolism. This PDK4 can be overexpressed by stimulating PPARα, which regulates fatty acid oxidation. Thus, we hypothesise that, at 4°C, upregulating PPARα should increase viability of hiPSC-CMs and downregulating it should decrease viability. Therefore, hiPSCs were differentiated into cardiomyocytes (CMs) and purified from non-myocytes using puromycin selection, resulting in a CM population with 99% purity. Then, PPARα was targeted in the 99% pure CMs using small molecules, and viability was assessed over 7 days at both 4 °C and 37 °C in various serum-free culture media. TNNT2-FITC and Calcein-AM flow cytometry staining were used to evaluate CM purity and viability, respectively. Interestingly, hiPSC-derived CMs stored at 4 °C exhibited higher viability compared to those stored at 37 °C. Treatment with the PPARα agonist resulted in a modest yet consistent increase in viability, whereas the PPARα antagonist caused a decrease. Finally, this preliminary experiment requires further optimization to yield statistically significant results, which is a promising approach for non-cryogenic preservation of cardiomyocytes.

https://doi.org/10.1007/978-3-319-45457-3_10

https://doi.org/10.1111/acel.13800

Abstract No. P012

Perlecan-conjugated laminin fragment as a next-generation culture substrate for human pluripotent stem cells

Kohei Minobe[1], *Yasuhiro Shimizu*[1,2], *Kakeru Sugiyama*[1], *Taiko Kunieda*[1], *Philipp Boder*[3,*], *Yukimasa Taniguchi*[1,2], and *Kiyotoshi Sekiguchi*[1,2]

[1] Matrixome Inc., Osaka, Japan
[2] Institute for Protein Research, Osaka University, Japan
[3] Amsbio, Alkmaar, The Netherlands

*Presenting author

Introduction: Cells in our body need to attach to the basement membrane, which mainly consists of laminin, collagen IV, nidogen and heparan sulfate (HS) proteoglycan (e.g. perlecan). Laminin and perlecan are key components to provide essential cues for stem cell maintenance and function. Laminin binds integrins to regulate cell survival, proliferation, and differentiation, while perlecan captures growth factors via HS chains attached to its N-terminal perlecan domain I (PDI), modulating growth factors availability to cell surface receptors.

Aim: Given the cooperative signalling between integrins and growth factor receptors, a culture substrate integrating both laminin's adhesive properties and perlecan's growth factor-binding capacity is highly desirable.

Methods: We engineered a recombinant laminin E8 fragment conjugated with perlecan's HS-rich PDI domain, which was produced in CHO-S cells and purified via a series of chromatographic steps.

Results: The resulting chimeric protein, named perLAM, fully retains both the integrin-binding activity of laminin and the growth factor-binding activity of perlecan.

Conclusion: This study highlights the use of perLAM as a "next-generation culture substrate" for stem cell culture & differentiation in regenerative medicine & tissue engineering using ES/iPS cells.

Abstract No. P013

Directed differentiation of pig pluripotent stem cells to definitive endoderm

Ekin Gökce Cicek[1,*], *Kaiyuan Yang*[1], *Elisabeth Kemter*[2], *Ejona Rusha*[3], *Eckhard Wolf*[2], *Carolin Daniel*[4], *Matthias Hebrok*[5], and *Heiko Lickert*[1]

[1] Institute of Diabetes and Regeneration, Helmholtz Munich
[2] Gene Center and Center for Innovative Medical Models (CiMM), LMU, Munich
[3] iPSC Core Facility, Helmholtz Munich
[4] Helmholtz Diabetes Center, RU Type 1 Diabetes Immunology
[5] Institute for Diabetes and Organoid Technology (IDOT)

*Presenting author

Directed differentiation of human pluripotent stem cells (hPSCs) into insulin-producing β-like cells offers promise for diabetes cell replacement therapy by overcoming donor scarcity. While allogeneic hPSC derived islet-like organoids have reached Phase I/II clinical trials, current protocols still yield heterogeneous populations, including proliferative progenitors or off-target cells, raising safety concerns. To address these limitations, we are developing a pig pluripotent stem cell (PSC) platform as a more physiologically relevant and translationally viable preclinical model. The pig model enables complementary in vitro and in vivo analyses of pancreatic development. Pig pancreas formation mirrors human morphogenesis and gene expression dynamics, providing sufficient time for studying early lineage transitions. We first optimized chemically defined porcine PSC maintenance protocols by systematically testing ESC media (Gao et al., 2019; Kinoshita et al., 2021) with different levels of Activin A, FGF2, LIF, and XAV939. This identified a formulation enabling robust feeder-free pluripotency maintenance, evidenced by >95% OCT4 and SOX2 co-expressing pPSCs. We further adapted human PSC protocols (Velazco-Cruz et al., 2019; Nair et al., 2019) to differentiate porcine PSCs toward definitive endoderm (DE) using Activin A and CHIR99021. We found that cytokine dose and timing critically affect differentiation efficiency, and we optimized conditions to achieve efficient DE commitment, with >90% co-expression of FOXA2 and SOX17. Overall, we have established a fully chemically defined platform for porcine PSC maintenance and endoderm differentiation, offering a powerful system to study pancreatic lineage development and advance safer, clinically relevant diabetes therapies.

Abstract No. P014

Exploring the impact of telomere-related mutations on iPSC reprogramming in a model of short telomere related pulmonary fibrosis

Amy Duffy[1,*], *Xi Jing Teoh*[1], *Wan Lin Ng*[1,2], *Aoife Carolan*[1], *Anja Schweikert*[1], *Mari Ozaki*[1], and *Killian Hurley*[1]

[1] Royal College of Surgeons in Ireland, Dublin, Ireland
[2] Beaumont Hospital, Dublin, Ireland

*Presenting author

Pulmonary Fibrosis (PF) is an interstitial lung disease characterised by lung tissue scarring, respiratory failure and death at a median of 3-5 years after diagnosis.

Telomeres are repetitive DNA sequences at chromosome ends that protect against degradation. Mutations in telomere-related genes (TRG) such as TERT, RTEL1, and PARN can cause telomere length shortening, and have been implicated in early-onset PF.

Induced pluripotent stem cells (iPSCs) are derived by reprogramming somatic cells providing patient specific in vitro models of type 2 alveolar epithelial (AT2) cells. iPSC reprogramming is expected to lengthen telomeres to resemble the telomere length (TL) in embryonic stem cells.

This study aimed to characterise TL and DNA damage in iPSC from patients with TRG mutations during reprogramming.

Peripheral blood mononuclear cells (PBMCs) from patients with short telomeres (characterised by Flow-FISH) and a TRG mutation were reprogrammed using Sendai virus. RNA and DNA samples were collected from the PBMCs of patients and non-diseased controls and from iPSCs at key passage (P) numbers. TL was measured via qPCR, telomere-related genes, pro-fibrotic, proliferation, and apoptotic gene expression was measured via RT-qPCR.

Our data shows that TL in both non-diseased control iPSC and iPSC derived from patients with an RTEL1 mutation, or a PARN mutation lengthen when compared to their parental PBMCs ad are comparable with each other. TL increase between P5 and P25 and stabilise by P30.

This is the first time that the role of TRG mutations on TL, and DNA damage during iPSC reprogramming has been investigated. Our data suggest that TL increases in RTEL1- and PARN-mutant iPSCs compared to their parental PBMCs, and the expression of telomere-related genes (TRGs), as well as fibrotic, proliferative, and apoptotic markers, remains comparable to non-disease controls. Notably, P53 and RTEL1 expression were elevated in TRG iPSCs.

Abstract No. P015

Serum-free differentiation and cultivation of human PSCs into endothelial cells using a closed semiautomated cultivation platform

*Kathrin Godthardt**, *Claudia Schreiner*, *Daria Sokoliuk*, and *Sebastian Knöbel*

Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach

*Presenting author

Background & Aim: The vascular system, lined by endothelial cells (ECs), provides a barrier to tissue and influences blood homeostasis. Among other processes endothelial cells are involved in neovascularization, which is essential for the growth and metastasis of tumors. In this context ECs transport nutrients and remove metabolic waste from tumor cells.

To understand and influence these interactions in more detail as well as to engineer vessels and organ grafts, large amounts of ECs are required. Pluripotent stem cell derived endothelial cells (PSC-ECs) can be produced in unlimited number without ethical concern, under standardized environment and thus provide an optimal source for studying processes mentioned above.

Methods: Endothelial differentiation of PSCs was performed within seven days using subsequently two different serum- and xeno-free cell culture media. Further cultivation for at least 14 days was performed under optimized xeno-free conditions. For scalable PSC-EC production we used the GMP compliant CliniMACS Prodigy® Adherent Cell Culture System.

Results: Resulting PSC-ECs revealed standard endothelial markers, showed DiI-acetylated LDL uptake and tube formation capacity.

Conclusion: Described is a standardized, scalable and closed system to produce functional PSC-ECs in serum- and xeno-free condition.

Abstract No. P016

Efficient and scalable 2D based differentiation of human PSCs into hematopoietic stem cells (HSCs) using a xeno- and serum-free cell culture medium

*Kathrin Godthardt**, *Tam Pham*, *Claudia Schreiner*, *Sofia Starkas*, *Sara de la Cruz Molina*, and *Sebastian Knöbel*

Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach

*Presenting author

Background & Aim: The differentiation of human induced pluripotent stem cells (iPSCs) into hematopoietic stem cells (HSCs) is since many years and important target for research and preclinical activities and creates big expectations for new therapeutic options in a range of hematopoietic disorders. This could range from autologous iPSC-HSCs to circumvent graft-versus-host-disease in recipients of imperfectly matched allogenic transplants, to autologous HSCs differentiated from genome-edited iPSC lines to treat patients by correcting genetic causes of blood diseases. Additionally iPSC-HSCs can always be used as an intermediate for further immune cell differentiation like macrophages, NK or T cells. To understand and influence this applications large amounts of functional iPSCs-HSCs are required. An efficient 2D based method to generate iPSC-HSCs is described here.

Methods: Hematopoietic differentiation of iPSCs was performed in a 2D approach within twelve days using subsequently three different optimized serum- and xeno-free cell culture media. For scalable iPSC-HSC production we used the GMP compliant CliniMACS Prodigy® Adherent Cell Culture System. The functionality of the generated HSCs was assessed in semi-solid cultures designed specifically to support erythroid and myeloid colony growth. Furthermore Lymphoid differentiation potential was assessed within an optimized differentiation process for iPSC derived NK cells.

Results: Resulting iPSC-HSCs revealed standard hematopoietic markers and showed myeloid as well as lymphoid differentiation potential in vitro.

Conclusion: Described is a highly efficient, scalable and closed system to produce functional iPSC-HSCs in serum- and xeno-free condition.

Abstract No. P017

Hepatocyte-like cells derived from alpha-1-antitrypsin deficiency-patient iPSCs reproduce disease-associated pathway dysregulation and react to potential drugs

Nina Graffmann[1,*], *Rabea Hokamp*[1], *Christiane Lörch*[1], *Malin Fromme*[2], *Nurdan Güldiken*[2], *Katharina Remih*[2], *Wasco Wruck*[1], *Pavel Strnad*[2], and *James Adjaye*[1,3]

[1] Institute for Stem Cell Research and Regenerative Medicine, University Düsseldorf
[2] Department of Internal Medicine III, RWTH Aachen
[3] EGA Institute for Women`s Health, Zayed Center for Research into Rare Diseases in Children (ZGR), London, UK

*Presenting author

The serine protease inhibitor (Pi) alpha-1-antitrypsin (AAT), produced and secreted from hepatocytes, attenuates neutrophil elastase mediated inflammation in the lungs. Severe Alpha-1-antitrypsin deficiency (AATD) is in 95% of the cases caused by an autosomal-dominantly inherited mutation in the *SERPINA1* gene resulting in a homozygous Glu342Lys substitution (Z-AAT). This leads to miss-folding of AAT and accumulation of the Z-variant within the endoplasmic reticulum (ER) of hepatocytes. Besides a lung phenotype, AATD manifests as liver fibrosis/cirrhosis in adults with no approved treatment so far.

In this project, we collect and bank urine derived MSCs from AATD-patients, carrying the homozygous Pi*ZZ genotype, and reprogram them into iPSCs. To model AATD, these iPSCs are differentiated into hepatocyte-like cells (HLCs).

Healthy and patient derived iPSCs could be differentiated into HLCs as documented by immunofluorescence (IF) and qRT-PCR for hallmark hepatocyte markers. Healthy and diseased lines could be distinguished by comparing AAT and Z-AAT levels in IF. HLCs were treated with the potential therapeutics Carbamazepine (CBZ), SAHA, Kifunensine (KIF), and Cysteamine (CYS). Of these, CBZ and SAHA could significantly reduce Z-AAT polymers.

Transcriptome analysis of healthy and diseased HLCs demonstrated dysregulation of many disease-associated KEGG pathways such as proteasome, Ca-signalling and bile secretion. Treatment with SAHA or CBZ had major impact on signalling pathways such as MAPK- and cAMP-signalling. In addition, SAHA treatment interfered with metabolism associated KEGG pathways reducing e.g. drug-, cholesterol-, or fatty acid metabolism.

These results give an insight into the potential of patient-specific HLCs generated from iPSCs as a suiting model for research on the individual genetical and molecular background of AATD. We will further exploit its functionality as a drug testing system and to increase our understanding of ZAAT polymer formation.

Abstract No. P018

Inter- and intra-donor variability of iPSC generated from human skin biopsies via an accelerated and validated pipeline using non-integrative RNA-based reprogramming

Diana Haberhausen[1,2,*], *Hannah Johannsen*[1], *Andreas Bosio*[1], and *Sebastian Knöbel*[1]

[1] Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
[2] UKB Universitätsklinikum Bonn

*Presenting author

Induced pluripotent stem cells (iPSCs) with their potential to differentiate into countless different cell types hold great potential for allogenic as well as autologous cellular therapy. However, conventional methods for iPSC generation often involve genomic integration of reprogramming vectors, thereby posing safety concerns that limit their clinical applicability. Moreover, established protocols are frequently time-consuming and necessitate labor-intensive screening procedures to exclude undesired vector integration. In contrast, transient mRNA-based reprogramming offers a non-integrative, rapid, and highly efficient alternative. Here we present a fast and reliable protocol to generate cryopreserved, quality-controlled, monoclonal iPSC lines from human skin biopsies. Fibroblasts were isolated from abdominal skin and minimally expanded before being reprogrammed via repeated mRNA transfections. iPSC colonies formed efficiently and were expanded into stable monoclonal lines. After seven weeks, iPSC lines were ready for banking, molecular characterization and further differentiation, minimizing time in culture and therefore the risk for cultivation induced aberrations. Thus, we generated a dataset comprising 78 iPSC lines derived from 10 donors (age 49-81). All tested lines showed characteristic morphology, pluripotency-associated marker expression and successfully differentiated into the three germ layers. While phenotypically all iPSC lines revealed low intra- and inter-donor variability, the verification of genetic integrity (digital PCR/SNP-based CNV analysis) and the assessment of oncogenic risk profile (whole exome sequencing) was more selective, highlighting the need for comprehensive quality control of newly generated iPSC lines. To address this, we implemented a systematic quality control pipeline for iPSC lines designated for downstream applications, that complies with ISSCR standards. In conclusion, the described protocol allows for efficient, reliable and integration-free generation of high-quality monoclonal iPSC lines from human skin punch biopsies within only seven weeks, thus facilitating individualized iPSC-based research and therapeutic development.

Abstract No. P019

Advanced hPSC bioprocessing via 3D inoculation from cryopreserved cell stocks

Carlos Alberto Hernandez Bautista[1,*], *Nils Kriedemann*[2], *Kevin Cyrys*[1], *Hugo Metzger*[1], *Annika Franke*[1], and *Jana Teske*[1]

[1] Hannover Medical School (MHH)
[2] Novo Nordisk, Copenhagen, Denmark

*Presenting author

Human pluripotent stem cells (hPSCs) have great potential for revolutionizing regenerative medicine. Despite recent progress in early clinical applications, differentiation and production of hPSC-progenies has been mainly studied in laboratory scale adherence-dependent (2D) cultures. This approach has substantial limitations regarding bioprocess efficiency, control and scalability for cell production in clinically relevant quality and quantities. For overcoming these, we have established different 3D suspension culture platforms, focusing on fully controlled stirred tank bioreactors (STBRs). By leveraging the application of STBRs, the mass production of 5.25^9 pluripotent hPS cells in a 150 mL working scale has been achieved within a 7-day process, resulting into a 70-fold expansion of the inoculated cell density. Nevertheless, pre-expansion of cryopreserved hPSC stocks via poorly controlled 2D culture was previously required before STBRs inoculation for 3D cultivation, creating challenges for good manufacturing practice (GMP)-compliant bioprocess development. Here we present a novel approach enabling the “high-density seeds” (HDS) for 3D bioprocess inoculation directly from cryopreserved hPSC stocks, thereby overcoming the need of any 2D culture. However, our novel strategy critically depends on the efficient recovery of viable hPSCs from cryopreservation followed by the impact of stirring shear stress in suspension. We have thus established screening assays for testing the efficiency of different “cell recovery reagents” with the HDS approach and will present solutions enabling efficient hPSC recovery and aggregation particularly during the critical phase post-inoculation. The resulting cultivation strategy is notably compatible with subsequent serial passaging of hPSCs combined with process upscaling, while all relevant hPSC characteristics such as exponential cell growth, pluripotency-associated marker expression, genomic stability and differentiation potential are maintained. Moreover, when combined with a directed differentiation protocol, the successful mass production of bona fide hPSC-cardiomyocytes in suspension culture has been demonstrated, fulfilling future clinical applications and high throughput screening platforms for drug testing as well.

Abstract No. P020

AI-based live imaging of a scaled-up production of human induced pluripotent stem cell-derived cardiomyocytes for in vitro test systems

Sabrina Herrmann[1,2,*], *Dominic Bauer*[2], *Ralf Kettenhofen*[2], *Krithika Hariharan*[2], *Julia C. Neubauer*[2], and *Heiko Zimmermann*[1,2]

[1] University of Saarland, Saarbrücken
[2] Fraunhofer IBMT, Sulzbach

*Presenting author

Background –Just recently, the U.S. Food and Drug Administration (FDA) has made the decision to reduce the use of animal testing by replacing it with adequate human-based *in vitro* models to produce monoclonal antibodies, among other objectives. Tremendous effort was put into the development of such systems by either the use of human primary or induced pluripotent stem cell (iPSC)-derived organ-specific cells. With a focus on the heart, the challenges with the *in vitro* culture of primary cardiomyocytes (CMs) include fast dedifferentiation, insufficient quantity, and inconsistent quality. Using standardized, quality-controlled human iPSC-CMs (hiPSC-CMs), which have already been validated for use in safety pharmacology, these issues can be overcome. Furthermore, implementing New Approach Methodologies (NAMs) has the potential to enhance these processes.

Results – We were able to scale up the production of hiPSC-derived CMs in a standardized and reproducible manner for the generation of cardiac in vitro models. Using a stirred bioreactor system, we yielded two billion hiPSC-CMs within eight days. During differentiation, AI-based live imaging was used to assess the quality of the process. Ultimately, the quality and high purity of the cells were confirmed after differentiation by the high expression level of cardiac troponin T, reaching up to 99%.

Conclusion – Substantial, quality-controlled batches of hiPSC-CMs were generated by scaling up the hiPSC-CM production. AI-based live imaging can be used to determine the fate of differentiation early on. The successfully differentiated cells have great potential for setting up standardized cardiac *in vitro* models. We demonstrated the functionality of our hiPSC-CMs, which makes them an even better candidate for setting up test systems for tissue engineering and large-scale drug screening.

Abstract No. P021

Bioprinted 3D pancreatic ductal tubes: A physiologically relevant model for studying ductal biology, disease mechanisms, and regenerative medicine

Fatemeh Navaee, *Majid Halvaei*, *Meike Hohwieler**, *Markus Breunig*, and *Alexander Kleger*

Institute of Molecular Oncology and Stem Cell Biology, University Ulm

*Presenting author

Introduction: Understanding pancreatic ductal biology is essential for studying pancreatic development, disease progression, and regenerative medicine. Traditional organoid cultures provide valuable insights but lack the precise spatial organization and structural fidelity of native pancreatic ducts. To address this limitation, we developed a novel 3D in vitro model of pancreatic ductal tubes using bioprinting technology.

Methods: We utilized an optimized bioprinting protocol to precisely deposit pancreatic progenitor (PP) cells, derived from human pluripotent stem cells, into a solidified Ultimatrix substrate. The printed constructs were then differentiated into ductal cells using a high-efficiency induction protocol. Comparisons were made between the bioprinted ductal tubes and pancreatic duct-like organoids (PDLOs) using light-sheet microscopy, immunofluorescence staining, and single-cell RNA sequencing (scRNA-seq) to assess i.e. lumen integrity, ductal identity (MUC1, CFTR, CA2), and polarization (Col4A1, AcTUB). Additionally, we investigated the effects of oncogenic mutations on ductal morphology.

Results: Our bioprinted pancreatic ductal tubes demonstrated superior structural integrity, with well-defined luminal architecture and enhanced spatial organization compared to PDLOs. Immunostaining revealed increased expression of key ductal markers, indicating a more advanced state of maturation. scRNA-seq analysis further confirmed activation of key signaling pathways, and improved transcriptomic alignment with native pancreatic ductal cells.

Furthermore, we found that bioprinted ductal tubes exhibited more pronounced phenotypic responses to oncogenic KRAS and GNAS than conventional 2D and organoid cultures. They displayed altered tube morphology, increased proliferation, and disrupted differentiation patterns, suggesting that the bioprinted system more accurately captures disease-associated cellular behaviors.

Conclusion: This study establishes a bioprinting-based approach that enables the formation of anatomically relevant, hollow ductal structures closely mimicking the architecture and cellular organization of native pancreatic ducts, providing a superior platform for studying pancreatic function and disease. The ability to model oncogenic mutations highlights its potential for investigating pancreatic plasticity, dysplasia, and cancer progression.

Abstract No. P022

Investigating the role of externally applied cyclic shear stress in early differentiation within iPSC-derived embryoid bodies

Syeda Inaas[1,*], *Sophia Götz*[2], *Rene Krüger*[1], *Erik Noetzel Reiss*[2], and *Wolfgang Wagner*[1]

[1] Institute for Stem Cell Biology, Uniklinik RWTH Aachen
[2] IBI-2, Forschungszentrum Jülich

*Presenting author

Early germ lineage specification is coordinated by the interplay of biochemical and mechanical cues. Mechanical cues can be modulated by the stimulation of cell membrane and cytoskeleton through application of forces. In this study, we want to understand if cyclic shear stress impacts differentiation of iPS cell aggregates - so called embryoid bodies - toward the three germ layers (endoderm, mesoderm, and ectoderm). To this end, we employed a magnetic shear stress device to subject the embryoid bodies to cyclic shear stress with a frequency of 0.2 Hz and an amplitude of 5 mm for a duration of 5 days. After 5 days of shear stress exposure, it was observed that the strain decreased by about 3 times suggesting that the matrix undergoes remodelling and transmits the forces differently. It was observed that the embryoid bodies, which were embedded in geltrex, showed a similar morphology in the presence and absence of shear stress. On assessing the gene expression levels, the embryoid bodies exposed to shear stress depicted a slight increase in expression of mesodermal marker HAND1 alongside a slight decrease in the pluripotency marker OCT4 in comparison to those that were not exposed to shear stress. Preliminary analysis suggested that the tight junction marker ZO-1 in embryoid bodies was up-regulated by shear stress, potentially indicating an improvement in apical polarization. RNA sequencing revealed hardly any significant transcriptional changes emerging from the shear stress and these were not related to early differentiation. Taken together, our results suggest that mechanostimulation of embryoid bodies by cyclic shear stress might result in a moderate upregulation of some markers for mesodermal differentiation, but these effects were not consistently observed.

Friedland, F., Babu, S., Springer, R., Konrad, J., Herfs, Y., Gerlach, S., ... & Noetzel, E. (2022). ECM-transmitted shear stress induces apoptotic cell extrusion in early breast gland development. Frontiers in Cell and Developmental Biology, 10, 947430.

Abstract No. P023

Serum- and xeno-free fibroblasts derived from human iPS cells

Yajuan Jiang[1,*], *Sarkawt Hamad*[2], and *Kurt Pfannkuche*[3]

[1] Center for Physiology and Pathophysiology, University of Cologne, Medical Faculty and University Hospital, Cologne
[2] Institute for Neurophysiology, University of Cologne
[3] Marga-and-Walter-Boll-Laboratory for Cardiac Tissue Engineering, University of Cologne

*Presenting author

Cardiac fibroblasts (CFs), cardiomyocytes, endothelial cells, and vascular smooth muscle cells are the principal cellular components of the heart. CFs produce extracellular matrix (ECM) proteins such as collagens and fibronectin, forming a supportive scaffold for cardiac cells. They play a critical role in maintaining cardiac structure, function, biochemical signaling, and electromechanical signal transduction. However, the limited availability of primary human CFs remains a significant bottleneck in advancing cardiac research. Recent studies have demonstrated that CFs can be derived from human induced pluripotent stem cells (hiPSCs), offering an unlimited and renewable cell source. In this study, we analyze the differentiation of fibroblasts from mesodermal progenitors in order to compare their properties to fully functional cardiac fibroblasts.

We present a refined differentiation strategy that utilizes sequential Wnt signaling modulation to induce mesoderm formation from hiPSCs. Next, hiPSCs derived mesodermal cells were differentiated into fibroblasts using PKA and basic fibroblast growth factor stimulation. On day 10 of differentiation, flow cytometry was performed using the fibroblast markers Vimentin (VIM) and Collagen Type I Alpha 1 Chain (COL1A1). The flow cytometry analysis showed 89% for COL1A1 and 60% for VIM marker expression at day 10 differentiation. Immunostaining confirmed COL1A1 expression.

In conclusion, a two-step differentiation method, from hiPSC to mesodermal, and further mesodermal to fibroblast could be a feasible method to generate serum and xenogen-free hiPSC derived fibroblast.

Abstract No. P024

Comparing single-cell and clump passaging in iPSC cultures: Implications for genomic stability and standardization

*Qianren Jin**, *Claudia de Guidi*, *Magdalena Madej*, and *Pia Johansson*

Cell & Gene Therapy Core, Lund University, Sweden

*Presenting author

Human induced pluripotent stem cells (iPSCs) have become an important tool for biomedical research by enabling disease modeling and personalized medicine. However, maintaining their genomic integrity during extended in vitro culture remains a significant challenge. One key yet underexplored variable is the method of passaging. While single-cell passaging offers advantages in culture standardization and scalability, concerns persist regarding its potential to induce genomic instability over time. This study aims to compare single-cell passaging (TrypLE) and clump passaging (EDTA) under defined, xeno-free culture conditions.

We cultured six well established iPSC lines for 20 passages under each condition, in biological triplicates and with different operators to eliminate handling bias. Genetic stability will primarily be assessed with copy number variation (CNV) analysis using high-resolution molecular karyotyping (SNP-array).

We will also screen for potential p53 inactivation to evaluate whether one passing method favours the stability of this pathway.

Our study will provide critical insight into how passaging technique impacts iPSC genomic stability, informing guidelines for both routine stem cell handling and clinical-grade cell manufacturing. If single-cell passaging proves safe and stable, it could reinforce its adoption in automated workflows. Conversely, detection of significant instability with single-cell passaging would highlight the need for revised culture practices. Ultimately, our findings aim to enhance the reproducibility, safety, and scalability of iPSC-based research.

Abstract No. P025

Gene editing in hPSCs: The importance of post-editing quality controls

Pia Johansson[1,*], *Magdalena Madej*[1], *Katrin Neumann*[2], *Chiara Ambrosini*[3], *Sema Aygar*[4], *Tõnis Org*[5], *Laurent David*[6], *Laura Batlle-Morera*[7], *Lyn Healy*[8], *Harald Stachelscheid*[9], and *Sebastian Diecke*[10]

[1] Lunds Universitet, Sweden
[2] Dresden University of Technology, Dresden
[3] Human Technopole, Milano, Italy
[4] Harvard Medical School, Boston, US
[5] University of Tartu, Estonia
[6] Nantes Université, Inserm, Nantes, France
[7] Center for Genomic Regulation, Barcelona, Spain
[8] The Francis Crick Institute, London, UK
[9] BIH Core Unit pluri. Stem Cells and Organoids (CUSCO) at Charité - Universitätsmedizin Berlin
[10] Max Delbrück Center, Berlin

*Presenting author

Human pluripotent stem cells (hPSCs) have been facilitating advancements in disease modelling, drug discovery, and studies of human development since their discovery. Their application was even further broadened by the advent of CRISPR-based precision genome editing.

However, inherent challenges such as genomic instability, altered differentiation capacity, and potential unintended CRISPR-induced modifications and off-target effects necessitate stringent quality control measures that adhere to established standards. Establishing and adhering to these standards is crucial for aligning research and publication practices, enhancing reproducibility and validation of results, and maintaining scientific integrity across the hPSC field.

Here, we propose a structured workflow, based on data from leading European core facilities, for crucial quality controls in genome-edited hPSCs. This workflow highlights essential assays for assessing genomic integrity and functional characteristics, while also identifying less critical tests. Additionally, we present guidelines for reporting genome editing experiments, emphasizing the importance of selecting appropriate control cells and robust experimental design.

Abstract No. P026

Reducing variability in primate iPSCs to improve cross-species comparisons

*Antonia Keßler**, *Felix Pförtner*, *Ines Hellmann*, and *Wolfgang Enard*

LMU, Munich

*Presenting author

Cross-species comparisons are crucial for understanding evolution and human biology. As molecular phenotypes are highly cell-type specific, orthologous cell types across species are needed for comparative studies. However, due to ethical and practical constraints, access to these cell types is often limited. Somatic cell reprogramming into induced pluripotent stem cells (iPSCs) has revolutionized cross-species studies, enabling the generation of otherwise inaccessible cell types from multiple species. This allows to study evolutionary processes on a molecular level by generating and culturing iPSCs from different primate species *in vitro*.

The defining feature of any iPSC line is pluripotency, its ability to differentiate into all three germ layers. Yet, despite the fact that iPSC lines typically express the appropriate marker genes and display the expected colony morphology, they often differ substantially in their differentiation potential. This variability is influenced by technical factors like cell type of origin, reprogramming method and culture conditions. Such factors may also exacerbate species differences, reducing the comparability of iPSCs across species. In particular, part of the variability in the differentiation potential among iPSC lines is thought to be due to incomplete reprogramming of the epigenetic state.

To address this, we generate iPSC lines from both human and cynomolgus macaque cells using fibroblasts or urine-derived stem cells as the primary cell source. Cells are reprogrammed with a classic approach or with a recently developed protocol incorporating a transient-naïve treatment step, which has been shown to reduce epigenetic memory and bias in iPSC lines.

With improved reprogramming, culture conditions and larger biological replicates, we aim to identify the condition that results in the highest similarity in expression profiles between human and cynomolgus macaque iPSCs and thereby seek to enhance the power of species comparisons.

Buckberry, S., Liu, X., Poppe, D. et al. Transient naive reprogramming corrects hiPS cells functionally and epigenetically. Nature 620, 863–872 (2023). https://doi.org/10.1038/s41586-023-06424-7

Abstract No. P027

Differentiation and characterization of astrocyte and endothelial cells from induced pluripotent stem cells of Alzheimer's disease patients which show altered blood-brain barrier integrity

Sohana Islam Khan

Uniklinikum Halle (MLU) & University of Leipzig

Aim

This study aims to develop and analyze an in vitro human model using astrocytes & endothelial cell derived from iPSCs from Alzheimer's & non-Alzheimer patients. The model is designed to mimic disease-related cellular mechanisms and enhance our understanding of astrocyte & brain capillary endothelial-like cells (BCECs) development and Alzheimer's pathology by comparing with healthy individuals with AD patients.

Method

We employed iPSCs from healthy elderly donors (BIONi037-A), AD patients with the APOE4 risk variant (UKBi011-A; STBCi006-A), AD patients with CRISPR-modified APOE3, and APOE knockout iPSCs (BIONi037-A-1). These iPSCs were sequentially differentiated into neural stem cells (NSCs), neural progenitor cells (NPCs), and then directed into the astrocyte & BCECs lineage using a specific protocol. The resulting cells were characterized using immunofluorescence (IF), quantitative real-time PCR, flow cytometry, and blood-brain barrier (BBB) integrity. Furthermore, astrocytes were integrated into a BBB in vitro model by co-culturing them with iPSC-derived (BCECs), with barrier integrity assessed via transendothelial electrical resistance (TEER) and a sodium fluorescein assay (NAFL).

Result

Astrocytes & BCECs from five human iPSC lines were analyzed for differentiation morphology and protein expression via IF staining using specific markers. Stage-specific primers revealed significant differences between AD and control lines. Flow cytometry on 33-day mature astrocytes showed up to 85.9% GFAP-positive cells, with the (37-A) control line having the highest count. Our finding showed BBB integrity assessed by TEER indicated higher resistance in co-cultures than monocultures and observed significantly higher NAFL accumulation in the disease group compared to controls.

Conclusion

We developed an in vitro protocol to differentiate astrocytes & BCECs from human iPSCs from five cell lines, revealing that the APOE allele is crucial, with APOE4 in AD patients showing markedly different outcomes compared to APOE3 controls. NAFL assay revealed increased accumulation in disease iPSC lines versus controls.

Abstract No. P028

Application of single cell RNA sequencing to characterize transient cell populations and molecular signalling during respiratory differentiation of human pluripotent stem cells

Anna Kuleshova[1,2,*], *Laura von Schledorn*[1,2], *Nicole Cleve*[1,2], *Julia Dahlmann*[1,2], *Maike Kosanke*[1], *Oliver Dittrich-Breiholz*[1], *Ruth Olmer*[1,2], and *Ulrich Martin*[1,2]

[1] Hannover Medical School (MHH)
[2] German Center for Lung Research (DZL), Hannover

*Presenting author

Our research group developed a protocol to differentiate human induced pluripotent stem cells (hiPSCs) into mature proximal and distal respiratory epithelia. While the protocol generally succeeds, further optimization is needed to improve differentiation efficiency and robustness. Although key signaling pathways in lung development have been identified, the optimal timing of their modulation remains to be defined, and the roles of additional pathways are still unclear. Moreover, the influence of cell cycle dynamics, epigenetic features of individual lines, and paracrine factors from non-target cell types on differentiation outcomes is yet to be fully understood.

To address these questions and guide protocol refinement, we investigated the cellular composition and molecular signaling of cells differentiated using our established approach. By deliberately selecting an inefficient differentiation scenario (~20% NKX2.1-positive cells), we aimed to identify dysregulated pathways and contaminating populations. Single-cell RNA sequencing (scRNA-seq) revealed two main populations: NKX2.1-expressing lung progenitors and a CDX2-expressing contaminating population, likely representing posterior endoderm derivatives. Further characterization identified additional cell populations expressing markers of pancreatic, intestinal, neuroendocrine, proliferative and apoptotic lineages, providing critical insights into culture heterogeneity. These scRNA-seq findings establish an essential basis for understanding off-target differentiation and improving protocol outcomes.

Based on these insights, targeted protocol modifications were introduced, resulting in a significant increase in the proportion of NKX2.1-expressing cells. Bulk RNA sequencing of cells differentiated under standard and modified protocols identified candidate signaling pathways, which are currently being validated *in vitro*.

Abstract No. P029

Novel isthmic-like neural progenitors with high capacity for mid-hindbrain boundary neuron generation in vitro and in vivo

Sarah Lang[1,*], *Ceren Duman Thier*[2], *Sascha Conic*[1], *Nils Wörner*[1], *Hannah Monyer*[2], *Andreas Trumpp*[1], and *Marc-Christian Thier*[1]

[1] German Cancer Research Center - HI-STEM, Heidelberg
[2] University Hospital Heidelberg

*Presenting author

The isthmic organizer (IsO) is a conserved signaling center at the mid-hindbrain boundary (MHB), precisely orchestrating regional identities of mid- and hindbrain structures of the developing vertebrate brain. Previously, we developed a direct reprogramming strategy to generate induced Neural Plate Border Stem Cells (iNBSCs) from human somatic cells, bypassing pluripotency and acquiring an anterior hindbrain identity [1]. Building on this, we established a protocol to derive novel isthmic region-like neural progenitors (istNPCs), a clonal and stable progenitor population, that closely mimics the isthmus and can be expanded under xeno-free conditions.

istNPCs exhibit robust self-renewal, clonal expansion, and long-term stability, while expressing key isthmic markers including GBX2, FGF8, EN1, and PAX2. We demonstrated the compatibility of our *in vitro* system with its *in vivo* counterpart by isolating the IsO from E8 mouse embryos and maintaining it in istNPC culture conditions. Moreover, istNPCs can be patterned toward the anterior MHB, reflecting their early neuroepithelial identity and developmental plasticity, with the ability to recapitulate both anterior and posterior regional identities of the isthmus.

Consistent with their regional identity, istNPCs can be efficiently differentiated into MHB neuronal subtypes, including dopaminergic and serotonergic neurons, both *in vitro* and *in vivo*. Functional maturation was confirmed by mass spectrometry detecting dopamine, serotonin, and their metabolic derivatives in culture.

In summary, the direct reprogramming of human somatic cells into stable, clonal isthmic neural progenitors not only offers a powerful model to study early human brain development but also represents a promising step toward the rapid and efficient generation of isthmus-derived neurons, such as dopaminergic neurons for their potential clinical applications in cell replacement therapies.

[1] Thier, M.C. et al. (2019). Identification of Embryonic Neural Plate Border Stem Cells and Their Generation by Direct Reprogramming from Adult Human Blood Cells. Cell Stem Cell 24(1):166–182.e13

Abstract No. P030

Modelling the molecular basis of human speech evolution

*Manqi Liang**, *Johanna Geuder*, and *Wolfgang Enard*

Ludwig Maximilian University, Munich

*Presenting author

Heterozygous loss-of-function mutations in the transcription factor FOXP2 lead to speech and language disorder. Two human-specific amino acid substitutions (T302N and N324S) occurred in the conserved FOXP2 protein after the human-chimpanzee split, suggesting a role of FOXP2 in the evolution of speech. Mouse models (Foxp2hum) with these human-specific substitutions showed enhanced synaptic plasticity in the form of stronger long-term depression (LTD) in the medium spiny neurons (MSN) of the striatum, decreased tissue dopamine levels, enhanced performance during certain learning tasks, and altered gene expression of striatal neuronal pathways during learning. To understand the molecular basis of speech development and evolution, we use homology-directed CRISPR-cas9 genome editing to create the two amino acid substitutions, which generates a “chimpanized” version of FOXP2 in human iPSC cell line. Additionally, we generate mouse iPSCs from Foxp2hum and WT mice. We will differentiate these cells into MSNs and subsequently perform RNA-seq to investigate to what extent human and mouse *in vitro* MSNs can recapitulate the differential expression found in the striatum of Foxp2hum mouse model.

Abstract No. P031

From research to clinical manufacturing: the reliability of bFGF in supporting iPSCs growth, pluripotency and differentiation

Marcus Bunn[1], *Kanika Singh*[2], *Isha Dey*[2], *Lauren Sangenario*[1], *Matt Dallas*[1], *Suchitra Sajja*[2], *Yeri Song*[1], *Claire Ruzicka*[1], *Christine Lansche*[3,*], and *Katie Taylor*[1]

[1] Thermo Fisher Scientific, Frederick, US
[2] Thermo Fisher Scientific, Bangalore, India
[3] Thermo Fisher Scientific, Darmstadt

*Presenting author

This study demonstrates the functional equivalence of three Gibco PeproTech basic fibroblast growth factor (bFGF) grades (research use only (RUO), animal origin–free (AOF), and PeproGMP proteins) in supporting induced pluripotent stem cell (iPSC) growth, pluripotency, and differentiation. Human dermal fibroblast-derived iPSCs, expanded across multiple passages in media supplemented with each bFGF grade, exhibited consistent growth, morphology, pluripotency, genetic stability, and differentiation potential. This consistency across bFGF grades ensures reliable performance in iPSC culture.

Abstract No. P032

Cryopolymer hydrogels as scaffolds for engineered heart tissues in vitro

Ziwei Lu[1,2,*], *Katharina Schwab*[3], *Sarkawt Hamad*[1,2], *Annette Schmidt*[3], and *Kurt Pfannkuche*[1,2]

[1] Center for Physiology and Pathophysiology, Institute for Neurophysiology, University of Cologne
[2] Marga-and-Walter-Boll-Laboratory for Cardiac Tissue Engineering, University of Cologne
[3] Department of Chemistry, University of Cologne

*Presenting author

For the *in vitro* culture of cells on stiff substrates such as polystyrene, there is evidence that passive mechanical substrate characteristics have negative effects on the physiology of various cell types. The mechanical priming of cells, which is caused by stiff culture substrates, is referred to as “stiff priming” and describes the alteration of the transcriptome and physiological behavior of the cells. Moreover, 2D cell culture environments do not possess the 3D structure of extracellular matrix (ECM) as found in native tissue. ECM is an extremely important component of the cell’s microenvironment.

The cardiac ECM with its comparable rigid but still elastic properties are of key importance to mimic cardiac tissue *in vitro*. At the same time, growing engineered tissues that develop this complex matrix *de novo* could turn out quite time consuming and has not been fully achieved so far. Therefore, we employ an engineered polyacrylamide (PAAm) hydrogel whose elastic modulus can be adjusted to match that of adult cardiac tissue.

We hypothesise that spherical PAAm cryopolymers (“cryobeads”) are a suitable extracellular matrix (ECM) replacement for generating functional cardiac microtissues for *in vitro* pharmacology and toxicology studies. PAAm cryobeads enable the expansion of human induced pluripotent stem (iPS) cells under pluripotency-maintaining conditions and support their directed differentiation into cardiomyocytes. We hypothesise that the elastic properties of the PAAm matrix facilitate the structural and electrophysiological maturation of CMs in a shorter timeframe than engineered tissues or complex organoid models, enabling long-term cultures of human iPS-CMs.

Two experimental strategies are presented: Expansion of human induced pluripotent stem cells and differentiation into cardiomyocytes on cryobeads, and seeding of pre-differentiated cardiomyocytes. Both strategies resulted in successful culture of human iPS-CMs for at least 20 days while maintaining their contractile function.

Key words: Cryopolymer hydrogel; Human induced pluripotent stem cells; cardiomyocytes

Pieroth, S, Heras-Bautista, C, Hamad, S, et al. Poly(acrylamide) Spheroids with Tunable Elasticity for Scalable Cell Culture Applications MACROMOL CHEM PHYS. 2022; 2200246.

Abstract No. P033

The landscape of membrane-less organelles in early human differentiation

Gizem Mansour[1,2,*], *Ali Oğuz Can*[1,3], *Andres Gonzalez-Suarez*[1,2], *Shruthi Buddharaju*[1,2], *Kenji Schorpp*[4], *Kamyar Hadian*[4], *Fabian J. Theis*[3], and *Nicolas Battich*[1,2]

[1] Helmholtz Pioneer Campus, Munich
[2] Institute of Epigenetics and Stem Cells, Helmholtz Munich
[3] Institute of Computational Biology, HMGU, Munich
[4] Molecular Targets and Therapeutics Center HMGU, Munich

*Presenting author

Stem cell differentiation is governed by gene regulatory networks (GRNs), which orchestrate transcription, translation, and RNA turnover. These processes are closely associated with specialized, non-membrane-bound structures in the nucleus and cytoplasm, known as membrane-less organelles (MLOs). These include nucleoli, Cajal bodies (CBs), nuclear speckles (NS), paraspeckles (PS), polycomb bodies (Pc), and promyelocytic leukemia protein nuclear bodies (PML-NBs), each contributing to the regulation of gene expression and dynamically adapting to cellular states. Despite their importance, the specific roles of MLOs in mammalian stem cell differentiation—particularly during early germ layer formation—remain underexplored.

For example, PML-NBs display unique morphologies—'linear' or 'rosette'-like—in early human pluripotent stem cell (hPSC) differentiation, contrasting with their spherical forms in somatic cells. NS also undergo dynamic morphological changes both during differentiation and across the cell cycle. hPSCs initially show abundant NS, which progressively specialize as differentiation proceeds. However, it is still not fully understood whether the morphological dynamics of nuclear MLOs are associated with specific differentiation trajectories, cell cycle states, or a combination of both.

To address this, we aim to characterize the dynamics of MLOs during hPSC exit from pluripotency. We employ iterative indirect immunofluorescence imaging (4i), a multiplexed imaging technique, optimized for high-resolution confocal microscopy. This enables simultaneous visualization of multiple nuclear MLOs (nucleoli, PML-NBs, CBs, NS, PS, Pc), germ layer markers (e.g., NANOG, PAX6, GATA6, BRACHYURY), and cell cycle indicators (e.g., Ki67, EdU). We analyze this multiplexed data using CAMPA, an AI-based model for spatial phenotyping, to dissect the intricate relationship between MLO dynamics and differentiation.

Our findings reveal new insights into the spatial and temporal regulation of MLOs and their potential roles in GRN-driven lineage commitment during early human development.

John T. Butler et al. (2009). Changing nuclear landscape and unique PML structures during early epigenetic transitions of human embryonic stem cells. Journal of Cellular Biochemistry, 107(4), 609–621. https://doi.org/10.1002/jcb.22183

Bernhard A. Kramer et al. (2023). Iterative indirect immunofluorescence imaging (4i) on adherent cells and tissue sections. Bio-protocol, 13(13), e4712. https://doi.org/10.21769/BioProtoc.4712

Abstract No. P034

GMP-compliant iPS cell lines show efficient cardiac induction in a new differentiation platform featuring scalability and enhanced robustness

*Soraia Martins**, *Joëlle Dimmler*, *Melanie Hühne*, and *Boris Greber*

Catalent Düsseldorf GmbH

*Presenting author

Induced pluripotent stem cell (iPSC)-based therapies hold significant promise for treating conditions such as heart failure, but their clinical implementation is hindered by stringent quality and consistency standards under Good Manufacturing Practice (GMP) regulations. To address these challenges, Catalent has established a robust GMP-compliant workflow for iPSC generation using episomal reprogramming of eligible EU and US donors. The resulting iPSC lines consistently met rigorous release specifications, exhibited no critical cancer-associated gene mutations, and demonstrated a low global mutation burden, reflective of their neonatal origin from cord blood. Beyond the generation of this critical starting material, Catalent has been developing novel and improved manipulation platforms to facilitate translation to a GMP setting in a partnering model.

A critical achievement has been the development of a GMP-compatible workflow to differentiate HLA-homozygous iPSCs into cardiomyocytes (CMs). Initial improvements were achieved by co-stimulating WNT and BMP signaling pathways during cardiac mesoderm induction. However, true robustness was achieved by incorporating additional FGF and TGFβ pathway modulation and optimizing media changes. These modifications eliminated bulk cell density as a critical process parameter and enabled consecutive, high-efficiency differentiation runs. The resulting CMs matured over time, developing a ventricular identity and pronounced sarcomeric structure. The workflow is scalable and compatible with various 3D culture systems. Furthermore, single-cell dissociation followed by re-aggregation enhanced cell purity and homogeneity while maintaining chromosomal stability. To assess functionality of the iPSC-CMs, the contractility force was measured using our recently developed assay based on engineered heart tissue (EHT). iPSC-CMs demonstrated a robust and consistent contractile profile, confirming their physiological functionality and therapeutic potential.

In summary, this integrated, GMP-compliant iPSC and differentiation platform addresses key bottlenecks in iPSC-based cardiac therapies. By combining genetic stability, scalable production, and demonstrated functional performance, this approach enables a streamlined path to clinical applications for the treatment of myocardial infarction.

Abstract No. P035

Research on cardiomyocyte maturation through mechanical variation of cell environment

Beate-Sophie Nenninger[1,2,*], *Michael M. Gepp*[2], *Patricia Schoof*[3], *Ralf Kettenhofen*[2], *Heiko Zimmermann*[2], and *Julia C. Neubauer*[2]

[1] Saarland University, Saarbrücken
[2] Fraunhofer IBMT, Würzburg
[3] Heinrich-Heine-University, Düsseldorf

*Presenting author

Cardiomyocytes derived from human induced pluripotent stem cells (hiPSCs) are an ideal cell-based model to study cardiac function, toxicology and disease in vitro. Although these cells can be effectively generated in the laboratory, their maturation remains a significant limitation. Strategies to enhance maturation include electrical stimulation and combining cardiomyocytes with other cell types. Notably, cardiomyocytes are sensitive to their mechanical environment, which is vital for their behaviour like induction of mechanical stress, the secretion and arrangement of extracellular matrix (ECM) proteins or biomaterials with varying structures.

The goal of this study is to identify environmental conditions for enhanced maturation of hiPS-derived cardiomyocytes and enhance their functional properties for research and therapeutic applications. The hydrogel utilized is based on ultra-high viscous (UHV) alginate, derived from different algal species, characterised by high biocompatibility and tuneable properties. By modifying the crosslinking process and selecting different alginate compositions, the mechanical characteristics of the hydrogel can be finely tuned.

Advanced techniques such as nanoindentation allow for quantitative assessment of stiffness and viscoelastic behaviour at the cell-matrix interface. The resulting data will be integrated with established cell analyses, including confluence, beating frequency, and gene expression of hiPSC-derived cardiomyocytes, along with their mechanical properties assessed via nanoindentation. Our findings indicate that variations in crosslinking parameters significantly impact hydrogel stiffness, directly influencing cellular behaviour and cardiomyocyte maturation.

This comprehensive analysis establishes a clear correlation between specific mechanical parameters of the hydrogel and improved outcomes in cardiac cell culture, enhancing our understanding of mechanotransduction in cardiac cells and providing a framework for developing advanced in vitro models for disease modelling and therapeutic testing.

Abstract No. P036

UV-micropatterning for high-throughput generation of embryoid bodies

*Vera Nischalke**, *Guan Ying Tsai*, *Mohamed H. Elsafi Mabrouk*, *René Krüger*, *Ian O. Shum*, *Kira Zeevaert*, and *Wolfgang Wagner*

Institute of Stem Cell Biology, RWTH Aachen

*Presenting author

Embryoid bodies (EBs) are three-dimensional aggregates of pluripotent stem cells. Due to their inherent ability to self-organize and differentiate they are applied in drug screening and regenerative medicine. Nevertheless, current EB formation methods remain challenging to standardize and scale for automated cell culture. In a previous study, we demonstrated that geometrically confined colonies of induced pluripotent stem cells (iPSCs) spontaneously detach after 5–7 days, forming size-controlled 3D aggregates via micro-contact-printed vitronectin circles. Building on these findings, we have now developed an ultraviolet (UV)-micropatterning approach that further improves the efficiency and homogeneity of EB generation. This method involves first passivation of the substrate, then selective exposure of defined areas to UV light to degrade cell-repellent molecules. Finally, the irradiated regions are coated with vitronectin to enable cell adhesion followed by cell seeding and eventually self-detachment of colonies as EBs. When we compared UV-micropatterning EBs with conventional methods for EB formation such as U-bottom wells and the commercially available AggreWell™ system, the results revealed similar morphology and differentiation potential. The aggregates maintained pluripotency in iPSC-culture media and could be induced for multilineage differentiation towards endoderm, mesoderm, and ectoderm, as confirmed by lineage-specific marker expression. Ongoing work focuses on analyzing potential differences in extracellular matrix composition and how these factors may influence lineage specification. Overall, our results highlight the advantages of UV-micropatterning as a scalable, automatable method to generate homogeneous EBs, paving the way for further advancements in stem cell-based applications. Our method can provide a standardized tool to investigate self-organization and differentiation potential for quality control of iPSC lines, such as PluripotencyScreen.

Abstract No. P037

Deciphering the regulatory role of long non-coding RNA in pancreatic development and diabetes

Francesca Piazzalonga[1,*], *Sarah Merz*[1], *Cécile Julier*[2], *Sandra Heller*[1], and *Alexander Kleger*[1]

[1] Universitatsklinikum Ulm
[2] Universitè de Paris, France

*Presenting author

Gene regulatory networks play a vital role in pancreatic development and function. However, pathomechanisms associated with the non-coding genome are just starting to be discovered. Our recent work demonstrated that mutations in the ONECUT1 gene are responsible for a new neonatal diabetes syndrome in homozygous patients, whereas heterozygous carriers develop early-onset diabetes. Stage-specific characterization of in vitro differentiated human embryonic stem cells (hESCs) revealed that ONECUT1 plays an important role in transcriptional regulation during pancreatic progenitor formation and the initiation of the endocrine developmental program. Nonetheless, the complex regulation of ONECUT1 expression is only partly uncovered.

While investigating the ONECUT1 locus, we found a variant most strongly associated with T2D (rs2440374), located within the lncRNA RP11-209K10.2 (lncRNA-RP11), 11 kbp from ONECUT1. The GTEx Portal shows decreased lncRNA-RP11 expression in the pancreas in the presence of the T2D-associated allele. This SNP also disrupts a binding motif for transcription factor NKX2.2, essential for endocrine pancreas development and islet-cell fate. Therefore, we hypothesize a regulatory role for this non-coding region.

After deletion of the lncRNA-RP11 gene, hESCs were differentiated into the pancreatic endocrine lineage. Although pancreatic progenitor formation was not impaired, stem cell-derived islets generated from these progenitors exhibited slightly reduced number and function of beta-cells, however, we could not detect a significant loss in ONECUT1 expression.

Our omics data revealed transcription factor binding peaks in the promoter region of lncRNA-RP11. Since promoters of lncRNAs can also act as enhancer elements regulating neighbouring genes, we anticipate that the lncRNA-RP11 promoter might regulate ONECUT1. To test this, we deleted only the lncRNA-RP11 promoter. These hESCs will be differentiated into pancreatic endocrine cells to evaluate effects on ONECUT1 expression and lineage specification.

This characterization will provide insights into the role of the lncRNA-RP11 locus in endocrine development and pathophysiology, improving understanding of diabetes-related transcriptional regulation.

Abstract No. P038

The role of proteolytic and mechanical stress in genome-edited hiPSC-derived cardiomyocytes as a model of myofibrillar myopathy 6

*Isabelle Risse**, *Bernd K. Fleischmann*, and *Michael Hesse*

Institute of Physiology I, University Bonn

*Presenting author

To maintain protein homeostasis in all organs, especially in muscle tissue after usage, misfolded and damaged proteins must be continuously degraded. This process is supported by chaperone-associated selective autophagy (CASA). In the CASA-complex, the co-chaperone BAG3 plays a major role by contributing to the autophagic degradation of damaged sarcomere proteins in striated muscles. A heterozygous amino acid substitution at position 209 of the BAG3 protein impairs its function and causes severe skeletal muscle weakness, neuropathy, and restrictive cardiomyopathy, the pathomechanism of which is still poorly understood.

To model BAG3^{P209L} myofibrillar myopathy *in vitro*, we generated an isogenic series of genome-edited human induced pluripotent stem cell (hiPSC) lines with heterozygous and homozygous BAG3^{P209L}-mutations. To better understand the cardiac pathomechanism, which manifests as the dissolution of sarcomere structures and the formation of BAG3-containing protein aggregates, we differentiated hiPSC into spontaneously beating cardiomyocytes (CM). Surprisingly, no obvious differences in morphology, functionality, and expression of proteins in the protein control system were detected.

To induce the pathological phenotype, hiPSC-CMs were exposed to proteolytic and mechanical stress by incubation with the proteasome inhibitor bortezomib or by applying uniaxial stretch with a cell-stretcher. Interestingly, a massive formation of BAG3^{+} and p62^{+} aggregates was observed in the hiPSC-CMs of the BAG3^{P209L} heterozygous and homozygous lines after incubation with bortezomib. In addition, protein expression analysis revealed a blockage of the autophagic flux in these CMs. Applying uniaxial stretch led to a build-up of protein aggregates and sarcomere damage exclusively in the BAG3^{P209L}-CMs, underlining the essential role of BAG3 and CASA in maintaining proteostasis after muscle usage. These stress models are ideal for testing gene therapy approaches and substance screens for treating the cardiac phenotype of MFM6.

Abstract No. P039

iPSC-derived cardiomyocytes to investigate the role of OCaR2 in abnormal calcium signalling in cardiac arrhythmias

Kristin Rädecke[1,], Roger Ottenheijm[1], Vanessa Kirschner[1], Lisa Gölz[1], Michael Berlin[1], Juan Camacho Londoño[2], Michelle Malz[1], Axel Kraft[1], Amelie Obermeier[1], Arne Hansen[3], Alex Cornean[1], Christian Grimm[4], Christian Wahl-Schott[5], and Marc Freichel[1]*

[1] Institute of Pharmacology, Heidelberg University Hospital
[2] Pharmacology and Toxicology, CSL Behring Innovation GmbH, Marburg
[3] Institute of Experimental Pharmacology and Toxicology, Hamburg University Hospital
[4] Walther-Straub Institute of Pharmacology and Toxicology, LMU München
[5] Institute of Cardiovascular Physiology and Pathophysiology, LMU München

*Presenting author

Alterations in cardiomyocyte Ca^{2+} homeostasis are a key trigger for abnormal impulse generation by early or delayed afterdepolarizations, and fatal arrhythmias can result from abnormal release of Ca^{2+} from intracellular stores. In cardiomyocytes, nicotinic acid adenine dinucleotide phosphate (NAADP) is generated in response to catecholamine stimulation. Recently, a novel signalling pathway in cardiomyocytes reveals NAADP-mediated Ca^{2+} release from endo-lysosomes via two-pore channels (TPCs)[1]. Our lab identified Organellar Ca^{2+} Regulator proteins (OCaRs) as critical regulators of Ca^{2+} release from these acidic organelles[2]. In a mouse model of OCaR2-deficiency, isoproterenol and NAADP evoked intracellular Ca^{2+} oscillations in cardiomyocytes. Inhibition of NAADP suppressed these exaggerated Ca^{2+} signals, as well as inactivation of TPCs. Furthermore, ECG recordings in OCaR2-deficient mice revealed an increased frequency of ventricular extrasystoles and sudden cardiac death, which was prevented by the inactivation of TPCs.

While our findings establish a critical role for OCaR2 in regulating Ca^{2+} dynamics and arrhythmogenesis in mouse cardiomyocytes, its function in cardiac physiology in a human context has not been studied yet. We generated OCaR2$^{-/-}$and OCaR2$^{-/-}$/TPC2 $^{-/-}$ induced pluripotent stem (iPS) cell lines to find out if these mechanisms can be replicated in a human model system. The iPS cells were differentiated to both ventricular and atrial cardiomyocytes and Ca^{2+} transients were recorded under catecholaminergic stimulation at different time points. Not only will the role of OCaR2 in the pathogenesis of ventricular arrhythmias be explored, but also the possible impact of OCaR2 on atrial arrhythmias and atrial fibrillation can be investigated with these cells. Furthermore, the contribution of OCaR2 in Ca^{2+} signalling and arrhythmicity will be assessed at various time points reflecting distinct cardiomyocyte maturity. Ultimately, these insights may uncover pathomechanisms and novel molecular targets for the prevention and treatment of cardiac arrhythmias in humans.

1. Capel, R. A. et al. J Biol Chem 290, 30087–98 (2015).

2. Tsvilovskyy, V., Ottenheijm, R et al. J Clin Invest, 134 134: e169428. (2024).

Abstract No. P040

Synthetic peptide growth factors functionally equivalent to FGF2 and KGF

Kazuki Shigematsu[1,*], *Kosuke Minamihata*[1], and *Bärbel Icheln*[2]

[1] PeptiGrowth, Tokyo, Japan
[2] Active Bioscience GmbH, Hamburg

*Presenting author

Conventional growth factors (GFs) used in regenerative medicine face several challenges, including lot-to-lot variability, potential contamination with biological impurities, low stability, and high manufacturing costs. To address these issues, PeptiGrowth Inc. has developed synthetic peptides that replicate the functions of conventional GFs while overcoming these limitations.

In this poster presentation, we will feature 2 alternative peptide growth factors: PG-011, and PG-012, which are synthetic peptides for Fibroblast Growth Factor 2 (FGF2) and Keratinocyte Growth Factor (KGF), respectively.

PG-011 exhibits similar activity to recombinant FGF2 (rFGF2). PG-011 supported the maintenance of pluripotency in iPS cells to a comparable extent as rFGF2, as demonstrated by morphology, proliferation, and pluripotency marker expression. PG-011 remained stable in media for 4 days at 37°C, while the stability of rFGF2 diminished. PG-011 can also be applied for developing cultivated meat.

PG-012 demonstrates similar activity to recombinant KGF (rKGF). PG-012 achieved comparable differentiation efficacy to rKGF in the process of differentiating iPS cells into pancreatic beta cells.

Including these two, we have launched 12 synthetic peptide growth factors and plan to launch 2 more products, PDGF-AA and IL-15. We believe that our synthetic peptide growth factors will bring innovative advances to the fields of regenerative medicine and cell therapy.

Abstract No. P041

Comparative analysis of different TBK1-ALS mutations in patient-derived motor neurons to develop a novel screening platform

Jahnavi Srinidhi[1,2,*], *Babak Loghmani*[1], *Isabel Loss*[1,2], *David Brenner*[3], *Rüstem Yilmaz*[1], *Philipp Koch*[1,2], *Rosanna Parlato*[1], and *Jochen Weishaupt*[1,3]

[1] Universität Heidelberg
[2] Central Institute of Mental Health, Mannheim
[3] Universität Ulm

*Presenting author

Heterozygous mutations in the TANK-binding kinase 1 (TBK1) gene lead to ALS. TBK1 is a pleiotropic kinase regulating various cellular processes, including autophagy. It contains four domains: An N-terminal serine/threonine kinase domain (KD), a ubiquitin-like domain (ULD), and two coiled-coil domains (CCD1/ CCD2). TBK1 loss-of-function variants cause early protein truncation, leading to reduced kinase activity and/or impaired substrate binding, to autophagy adaptor proteins. This study had two major aims: 1) to characterize the cell-autonomous molecular effects of different TBK1 mutations in an ALS-vulnerable neuronal population, and 2) to identify specific phenotypic markers. We reprogrammed human iPSCs from peripheral blood mononuclear cells (PBMCs) of one ALS pre-manifest carrier of a heterozygous TBK1 splice-site mutation (c.1760+1G>C) and one ALS-affected family member with a heterozygous TBK1 frameshift mutation (c.78_79delAA/p.27Thrfs*2), differentiating them into motor neurons (MNs). We also included heterozygous and homozygous lines carrying the TBK1 missense mutation p.E696K, along with their isogenic control. To test whether this missense mutation impairs autophagy, we performed immunofluorescence analysis. In the homozygous MNs, we observed significant accumulation of p62-positive autophagosomes, LAMP2-positive lysosomes, and increased Galectin8-positive puncta indicative of damaged lysosomes. This suggests impaired lysosomal degradation. We characterized these puncta by size, number, and intensity, providing a potential readout for high-throughput drug screening and development of mutation-specific therapies. Additionally, we found increased frequency and length of primary cilia in TBK1 mutant MNs. These preliminary results suggest a role for TBK1 in regulating ciliogenesis. Given the interplay between cilia dynamics, autophagy, and the cell cycle, we aim to further investigate if TBK1 mutations disrupt these interconnected pathways. This emerging direction may uncover novel mechanisms through which TBK1 dysfunction contributes to ALS pathogenesis.

Abstract No. P042

Identification of general and tissue-specific mechanisms of Fabry disease using a patient-specific induced pluripotent stem cells model

Ferdinand Teichert[1,2,*], *Christian Schröter*[1,3], *Andreas Leffler*[4], *Robert Zweigerdt*[2], and *Micha Drukker*[1,5]

[1] Leiden Academic Centre for Drug Research (LACDR), Leiden University, The Netherlands
[2] Department of Cardiac, Thoracic, Transplantation and Vascular Surgery (HTTG), LEBAO, Medizinische Hochschule Hannover
[3] Max Planck Institute of Molecular Physiology, Dortmund
[4] Department of Anesthesiology and Intensive Care Medicine, Pain Management Service, Medizinische Hochschule Hannover
[5] Institute of Stem Cell Research, Helmholtz Munich

*Presenting author

Fabry disease is an X-linked disorder manifesting with progressive organ damage, especially in cardiovascular and nervous systems, inducing chronic pain among other symptoms. Caused by mutations in the galactosidase alpha (GLA) gene the resulting deficiency of the lysosomal enzyme α-galactosidase A leads to accumulation of glycosphingolipids (e.g., Gb3). Glycosphingolipids are proposed to modulate metabolic pathways contributing to autophagy and cellular decay, but the mechanisms of cellular damage are not fully understood, and no adequate *in vitro* models exist. Applying induced pluripotent stem cell (iPSC) technology, this project aims to establish advanced *in vitro* models of Fabry disease for investigating the pathophysiological mechanisms.

We have obtained and quality controlled previously established patient-derived iPSC lines for pluripotency associated markers expression via immunofluorescence staining and qPCR. The chromosomal integrity was confirmed via short-tandem repeat (STR) analysis and copy number variation (CNV), respectively. Fabry-specific mutations (1193_1196del [p.Glu398Glyfs*5]; 851T>C [p.Met284Thr]) and respective corrections in isogenic lines were validated by Sanger sequencing. The successful differentiation of these iPSC lines into beating cardiomyocytes and axial stem cells (AxSc) was achieved, promoting their envisioned co-culture. Moreover, the progressive differentiation of AxSc into sensory neurons (expressing required nociceptors) is ongoing, to enable the modeling of aspects of the cardiac and nervous manifestations of Fabry disease in a dish.

The model will be applied to measure functional and transcriptional changes in repose to pain-stress-like conditions resulting e.g. from chemical and thermal stimuli. Results will be compared between Fabry-derived vs. isogenic controls to identify common and cell type-specific pathophysiological effects. Molecular markers of fibrosis, autophagy, and inflammation will be analyzed to monitor aspects of disease progression.

Ultimately, the model may provide the basis for establishing new targeted therapies addressing neuropathic pain and cardiac dysfunction in Fabry patients and potentially promote studies on other chronic pain disorders as well.

Abstract No. P043

Salt-inducible kinases transduce mechanical forces into the specification of the pancreatic endocrine lineage

Chenglei Tian[1,*], *Adam Rump*[1,2], *Christine Ebeid*[2], *Anant Mamidi*[2], and *Henrik Semb*[1]

[1] Helmholtz Zentrum Munich
[2] University of Copenhagen, Denmark

*Presenting author

The extracellular matrix-F-actin-Yes-associated protein 1 (YAP1)-Notch mechanosignaling axis is a gatekeeper in the fate decisions of bi-potent pancreatic progenitors (bi-PPs). However, the link between F-actin dynamics and YAP1 activity remains poorly understood. Here, we identify salt-inducible kinases (SIKs) as mediators of F-actin-triggered changes in YAP1 activity. Interestingly, sodium chloride treatment promotes the differentiation of bi-PPs into NEUROG3+ endocrine progenitors (EPs) through enhanced SIK expression. Consistently, the pan-SIK inhibitor HG-9-09-01 (HG) inhibits latrunculin B (LatB)-induced EP differentiation via nuclear YAP1 accumulation. Unexpectedly, withdrawal of HG after a 12-h treatment increased SIK expression by a negative feedback mechanism, leading to significantly enhanced endocrinogenesis. Therefore, the combined treatment of bi-PPs with LatB and HG for 12 h boosted endocrinogenesis, ultimately leading to an increased number of beta cells. In summary, we identify SIKs as new transducers of mechanotransduction-triggered induction of pancreatic endocrine cell fates.

https://doi.org/10.1016/j.stemcr.2025.102444

Abstract No. P044

Pancreatic alpha and beta cell fate choice is directed by apical-basal polarity dynamics

Chenglei Tian[1,*], *Ulf Tiemann*[2], *Florian Hermann*[2], *Martin Proks*[2], *Emilie Skovgaard*[2], *Ivan Kulik*[1], *Yilin Di*[1], *Jakub Sedzinski*[2], and *Henrik Semb*[1]

[1] Helmholtz Center Munich
[2] University of Copenhagen, Denmark

*Presenting author

A central question in cell and developmental biology is how extracellular cues control the differentiation of multipotent progenitors in a dynamically changing niche. Here, we identify apical-basal polarity as the main regulator of the differentiation of multipotent pancreatic Neurogenin3+ endocrine progenitors (EPs) into the beta or alpha cell fates. We show that human EPs dynamically change their apical-basal polarity status. Whereas polarized EPs are predisposed to differentiate into beta cells rather than alpha cells, inhibiting apical-basal polarity selectively suppresses beta cell differentiation. Single-cell RNA sequencing and complementary mechanistic data demonstrate that apical-basal polarity in human EPs promotes beta cell specification via cyclic AMP (cAMP)/PKA-cAMP response element binding protein (CREB)-EGR1-mediated inhibition of ARX expression, while reduced cAMP levels in non-polarized human EPs maintain expression of ARX, leading to alpha cell differentiation. These findings identify the apical-basal polarity status of multipotent EPs as a critical epithelial feature that determines their fate into the alpha or beta cell lineages.

https://doi.org/10.1016/j.devcel.2025.02.008

Abstract No. P045

HERV-H activation disrupts endoderm formation via ZEB2-mediated inhibition of TGF-β/SMAD signaling

Vidya Padmanabhan Nair[1], *Hin-Man Tai*[1], *Goar Frishman*[1], *Wen Liang*[1], *Hengyuan Liu*[2], *Daniel F. Kaemena*[1], *Dmitrij Frishman*[2], and *Michelle Vincendeau*[1,2,*]

[1] Helmholtz Munich
[2] Technical University Munich

*Presenting author

Human endogenous retroviruses (HERVs) have emerged as critical regulators of early development, with specific groups influencing lineage commitment, differentiation, and cellular identity during embryogenesis. In this study, we investigated the functional role of HERV-H on germline differentiation using CRISPR-based approaches to modulate HERV-H expression. Bulk RNA sequencing revealed that HERV-H activation induces gene expression signatures characteristic of early human development. Upon directed differentiation into the three germ layers—ectoderm, mesoderm, and endoderm—HERV-H activation selectively impaired the formation of definitive endoderm, as demonstrated by reduced expression of canonical endodermal markers such as FOXA2 and SOX17. Whole-transcriptome analysis of endodermal cells revealed widespread dysregulation of genes involved in developmental and homeostatic signaling pathways essential for endoderm specification. Strikingly, HERV-H activation was associated with a strong upregulation of ZEB2, a transcriptional repressor known to inhibit endoderm differentiation, and a concomitant downregulation of SMAD2 and SMAD3, key components of TGF-β signaling. These findings suggest that aberrant HERV-H activity disrupts the transcriptional framework necessary for endodermal commitment, potentially by interfering with SMAD-dependent signaling and inducing inhibitory pathways. Altogether, our data uncover a previously unrecognized role for HERV-H in modulating cell fate decisions through coordinated regulation of transcriptional networks and signaling cascades during early human development.

Abstract No. P046

Rapid generation of oligodendrocyte progenitor cells from neural progenitor cells through 3D spheroid culture and small molecule modulation

Nils Wörner[1,*], *Minyoung Kim*[1], *Marcel Weinreich*[2,3], *Ceren Duman Thier*[1], *Hannah Monyer*[3], and *Marc-Christian Thier*[1]

[1] German Cancer Research Center - HI-STEM, Heidelberg
[2] German Cancer Research Center, Heidelberg
[3] University Hospital Heidelberg, Heidelberg

*Presenting author

Oligodendrocytes are the myelinating cells of the brain. They facilitate efficient signal conduction via myelination, while also playing a critical role in metabolic support and neural circuit modulation. Their loss or dysfunction contributes to various disorders, including leukodystrophies and neuroinflammatory conditions such as multiple sclerosis.

The study of oligodendrogenesis using current methodologies, such as human pluripotent stem cells, is limited due to the lengthy and low-yield differentiation protocols, which often require 75–100 days to generate mature oligodendrocytes.

We present a streamlined strategy for accelerating the production of oligodendrocyte progenitor cells (OPCs) from a specific type of neural progenitor cell: induced neural plate border stem cells (iNBSCs) (1).

Building on our previous finding that iNBSCs can efficiently generate oligodendrocytes in vivo following brief patterning with small molecules, we aimed to translate this capacity in vitro to enable the study of oligodendrogenesis. Ultimately, we identified the formation of 3D spheres as critical to improving the efficiency of OPC induction in vitro. For this, we developed 3D-printed moulds for PDMS casting to generate spheres of a homogenous size. These spheres were then exposed to a defined cocktail of small molecules to direct OPC specification. Notably, modulation of the FGF signaling pathway was essential for inducing cell migration out of the spheres and OPC fate upon subsequent plating. Preliminary data show that this combined approach yields ~38% OLIG2+/SOX10+ OPCs by day 30, with progression to pre-myelinating MBP+ oligodendrocytes by day 50 — shortening the timeline compared with conventional methods.

Our reproducible 3D system standardizes sphere formation, improving efficiency and homogeneity, with potential for straightforward scalability. These findings highlight the potential of iNBSCs in conjunction with 3D culture and FGF modulation to enhance OPC generation and pave the way for studying oligodendrogenesis and advancing translational remyelination strategies.

(1) Thier, Marc Christian, et al. "Identification of embryonic neural plate border stem cells and their generation by direct reprogramming from adult human blood cells." Cell stem cell 24.1 (2019): 166-182.

Abstract No. P047

Production of CAS9 expressing hiPSCs for genetic screen

Ferhat Ali Yaman[1,2,*], *Merrit Romeike*[1,2], *Fabian Bachinger*[1,2], *Dominika Dziedzicka*[3], and *Ludovic Vallier*[1,2]

[1] Berlin Institute of Health (BIH), Berlin
[2] Max Planck Institute for Molecular Genetics, Berlin
[3] Wellcome-MRC Cambridge Stem Cell Institute, Cambridge, UK

*Presenting author

Metabolic dysfunction-associated steatotic liver disease (MASLD) is a chronic disease linked to diabetes and characterized by lipid accumulation in hepatocytes. The disease progresses through stages of steatosis, hepatocyte injury, fibrosis, inflammation and cirrhosis, leading to liver failure or hepatocellular carcinoma. End stage of MASLD can only be treated with liver transplantation. Despite affecting approximately one-fourth of the global population, and with incidence rates expected to rise over the coming years, there are currently no approved drug to prevent lipid accumulation in the liver. Therefore, there is great interest to identify novel druggable targets to halt or to reverse MASLD progression.

To address this challenge, we aim to develop a scalable CRISPR-knockout screening strategy to identify regulators of lipid metabolism using human induced pluripotent stem cells (hiPSCs) forward programmed hepatocytes (FoP-Heps). However, stable expression of CAS9 remains difficult to achieve especially during differentiation thereby limiting the development of such screen. To overcome this, we have generated hiPSC lines expressing CAS9 under the control of different constitutive or tissue specific promoters. We then monitored transcript and protein levels during hepatocytes production and validated CAS9 activity. We also confirmed that gene targeting did not affect pluripotency or differentiation.

These engineered cell lines lay the groundwork for developing CRISPR screens to uncover novel regulators of lipid metabolism in MASLD, which could serve as new therapeutic targets. Of note, our gene expression strategy might be applied to different cell types and enable CRISPR knock out screens in a diversity of cell types.

P048 – P051: Stem cells in aging & disease (cancer stem cells)

P048 Gradient of Wnt signaling facilitates Mef2 heterogeneity and limits commitment of the developmental muscle progenitor pool
Josephine Bageritz

P049 Organ identity imprints non-linear aging of the hematopoietic system
Nina Henning

P050 Breast cancer stem cells induce IRF6 in stromal fibroblasts to upregulate stromagenesis
Jochen Maurer

P051 Delineating proteo-genomic signatures of human leukemic stem cells in AML at single-cell resolution
Tessa Schmachtel

Abstract No. P048

Gradient of Wnt signaling facilitates Mef2 heterogeneity and limits commitment of the developmental muscle progenitor pool

Laura Schütze, *Maria Jelena Musillo*, *Bilyana Popova*, *Natalja Engel*, *Verena Holzwarth*, *Milena Lucia Hild*, *Anna Geißdorf*, *Jennifer Nadine Groß*, *Ingrid Lohmann*, and *Josephine Bageritz*[*]

Heidelberg University

*Presenting author

During skeletal muscle development, the timing and extent of progenitor differentiation must be finely tuned to ensure proper tissue formation while maintaining a reservoir of undifferentiated cells for future regenerative needs. Myogenic commitment is driven by transcription factors like Mef2, while factors such as Zfh1 (ZEB1 in vertebrates) counteract this process to preserve progenitor identity. Yet, how these opposing cues are spatially coordinated within the developing niche remains poorly understood. Using the *Drosophila* indirect flight muscle (IFM) system, a powerful model with conserved parallels to vertebrate myogenesis, we uncover how spatially patterned Wnt/β-catenin signaling sculpts muscle progenitor (MP) fate decisions during a critical developmental window. We identify a dorsal subpopulation of IFM-MPs that exhibits low *Mef2* and high *Zfh1* expression during late larval stages. Spatial mapping, single-cell transcriptomics, and genetic perturbations reveal that graded Wnt/β-catenin activity maintains this less committed state by repressing *Mef2* in a dose-dependent manner. Moderate Wnt levels additionally sustain *Zfh1* expression. Disruption of Wnt signaling/β-catenin results in premature Mef2 upregulation and a loss of undifferentiated progenitors. We are currently investigating whether the size of this Mef2-low progenitor population influences the number or timing of adult muscle stem cell formation, offering a potential developmental link to long-term regenerative capacity. Notably, while Wnt/β-catenin signaling is classically associated with promoting myogenesis, our findings suggest that in this early spatial context, it instead supports a less committed progenitor state generating a subset of MPs with particularly low *Mef2* expression. This may represent a conserved strategy for regulating muscle stem cell formation across species.

Abstract No. P049

Organ identity imprints non-linear aging of the hematopoietic system

Nina Henning[1,*], *Giannoula Savvou*[1], *Max Pleyer*[1], and *Claudia Waskow*[1 2]

[1] Leibniz Institute on Aging (FLI), Jena
[2] Friedrich-Schiller University Jena

*Presenting author

Aging profoundly alters hematopoiesis, yet its impact varies significantly across organs. We observed a stark decline in lymphocytes in aged blood and thymus, while spleen and bone marrow maintained leukocyte populations comparable to young mice. Within the bone marrow, the primary site of hematopoiesis, we observed stable numbers for myeloid progenitors with age, an increase in platelet-biased hematopoietic stem cells, and preserved unbiased stem cell numbers. Intriguingly, functional analysis revealed that HSCs from aged mice can effectively repopulate their niche and the overall hematopoietic system in non-irradiated recipients, demonstrating maintained self-renewal and multi-lineage differentiation capacity under normal conditions. However, a lymphopoietic defect emerges in aged HSCs upon transplantation into irradiated recipients, mimicking the endothelial cell depletion observed in aged bone marrow niches. This highlights the critical influence of the aging microenvironment on the functional capacity of aged HSCs.

This organ-specific aging is driven by distinct local environments, with minimal overlap in key factors. Notably, proteases and matrix remodeling enzymes exhibited the highest organ-specific variance, exceeding that of pro-inflammatory cytokines. These organ-specific environmental cues correlate not only with the observed hematopoietic aging patterns but also with complex variations in cellular senescence across different tissues. Our findings underscore the critical role of the local organ microenvironment in shaping the aging trajectory of the hematopoietic system.

Abstract No. P050

Breast cancer stem cells induce IRF6 in stromal fibroblasts to upregulate stromagenesis

Harshini Muralidharan, *Thomas Hansen*, *Elmar Stickeler*, and *Jochen Maurer*[*]

University Hospital RWTH Aachen

*Presenting author

In the microenvironment of cancer stem cells (CSCs) we often find cancer-associated fibroblasts (CAFs). Here, we show the first in-depth analysis of the interaction between primary triple-negative breast cancer stem cells (BCSCs) with naive fibroblasts inducing a CAF phenotype. Using 2D coculture models with specific seeding ratios, we identified stromal fibroblast aggregation at the BCSC cluster periphery, and, on closer observation, the aggregated fibroblasts were found to encircle BCSC clusters in nematic organization. In addition, collagen type I and fibronectin accumulation were also found at the BCSC–stromal periphery. Sequencing of BCSC-encapsulating fibroblasts displayed the transformation of stromal fibroblasts to CAFs and the upregulation of fibrosis regulating genes of which the Interferon Regulatory Factor 6 (IRF6) was the most prominent. Loss of function experiments with IRF6 decreased fibroblast encapsulation around BCSC clusters in 2D co-cultures. In BCSC xenografts, fibroblast IRF6 over-expression led to an increase in stromal area and fibroblast density in tumors, in addition to a reduction in necrosis. Based on our findings, we propose that fibroblast IRF6 function is an important factor in the development of the stromal microenvironment and in sustaining the BCSC tumor niche.

Muralidharan H, Hansen T, Steinle A, Schumacher D, Stickeler E, Maurer J. Breast Cancer Stem Cells Upregulate IRF6 in Stromal Fibroblasts to Induce Stromagenesis. Cells. 2024 Aug 31;13(17):1466. doi: 10.3390/cells13171466. PMID: 39273037

Abstract No. P051

Delineating proteo-genomic signatures of human leukemic stem cells in AML at single-cell resolution

Tessa Schmachtel[1,*], *Weijia Yu*[1], *Sebastian Wolf*[1], *Elisa Donato*[2], *Marius Külp*[1], *Hana Komic*[3], *Malin S. Nilsson*[3], *Catia Simoes*[4], *Felipe Prosper*[4], *Adrien Jolly*[1], *Halvard Bonig*[5], *Bruno Paiva*[4], *Fredrik B. Thorén*[3], *Andreas Trumpp*[2], *Thomas Oellerich*[1], and *Michael Rieger*[1]

[1] Goethe University Frankfurt, University Hospital, Department of Medicine 2 - Hematology and Oncology, Frankfurt am Main
[2] German Cancer Research Center (DKFZ), Heidelberg
[3] TIMM Laboratory at Sahlgrenska Center for Cancer Research, Institute of Biomedicine, University of Gothenburg, Sweden
[4] Cancer Center Clínica Universidad de Navarra, Centro de Investigación Médica Aplicada (CIMA), Pamplona, Spain
[5] German Red Cross Blood Service Baden-Württemberg-Hessen, Frankfurt am Main

*Presenting author

Acute myeloid leukemia (AML) is characterized by cellular heterogeneity at an inter- and intra-patient level. Hierarchically organized differentiation trajectories and clonal evolution contribute to the heterogeneity of the disease. Leukemia-initiating stem cells (LSCs) are at the apex of the leukemic differentiation hierarchy feeding the pool of leukemic blasts. However, the phenotype of LSCs is highly variable between patients, and these rare cells share many molecular and functional features of hematopoietic stem and progenitor cells (HSPCs). Single cell analyses are required to resolve LSC heterogeneity, to study their molecular traits, and to identify molecular targets.

Therefore, we generated the largest atlas of human bone marrow stem and progenitor cells from 15 healthy donors covering young to old age groups, utilizing proteo-transcriptomic sequencing at a single cell level with the BD Rhapsody system. Over 62,000 HSPCs were analyzed for gene expression, surface marker profiles, differentiation trajectories, and age-related changes in hematopoietic fate decisions. Building on this reference dataset, we analyzed samples from 12 AML patients with DNMT3A-mutated AML—six presenting early relapse and six from patients showing long-term remission—using single-cell proteo-transcriptomic profiling. The AML samples were enriched for GPR56+ cells—a marker associated with LSCs—and data from over 39,000 single cells were projected onto our healthy reference dataset. This allowed us to identify molecular traits distinguishing LSCs from healthy HSCs influencing leukemic stem cell fate decisions, differentiation pathways and molecularly targetable pathways. By integrating transcriptomic and proteomic data, we aim to uncover LSC-specific behavior and to identify novel therapeutic targets.

P052 – P072: Tissue-derived stem cells & organoids

P052 Automated high-throughput generation and characterization of human neuromuscular organoids
Inês Afonso Martins

P053 Stem cell activation in the human stomach is spatially governed by a YAP1-DKK1 axis
Francesco Boccellato

P054 Role of dermal-epidermal junction topography in regulating epidermal stem cell fate
Adil Boolani

P055 Enhancing consistency in 3D spheroid engineering: A novel automated cell picking approach
Sofia Kuzmich

P056 Cellular plasticity and replenishment capacity in human brain organoids
*Christopher E*sk

P057 High fat diet affects the stromal niche signaling in the porcine intestine
Falk Janos Farkas

P058 TBR2 directs neuronal subunits switching in ATP-Dependent Chromatin Remodeling Complexes to regulate chromatin accessibility during Human Cortical Development
Catello Guida

P059 Scalable, defined cardiac organoids for drug discovery and non-cryogenic distribution
Sarkawt Hamad

P060 Pancreatic ductal organoids for basic developmental biology and drug screening applications
Christos Karampelias

P061 Modeling radiation-induced neurotoxicity in human cerebral organoids
Leon Kaysan

P062 Human otic Bioengineered Neuronal Organoids (oBENOs) mimic early inner ear development and physiology
Angeliki Koufali

P063 Modulation of ERK signaling dynamics by pharmacological intervention directs cell differentiation
Pavlina Markova

P064 Organoid assembloids reveal serotonergic regulation of human cortical development
Raquel Pérez Fernández

P065 CSF1R-dependent myeloid cells direct hepatocyte development in human and mouse models
Susanna Quach

P066 Self-organizing vascularization in human neuromuscular organoids drives functional maturation and regenerative capacity
Anna Rafiei Hashtchin

P067 Exploring the regional identity and chromatin landscape of human embryonic neural stem cells
Amelie Schurer

P068 Reproducible and efficient organoid development using stable and highly active synthetic peptide alternatives to BDNF, Noggin, and Wnt3a
Kazuki Shigematsu

P069 Label-free live cell imaging for spatiotemporal quantification of self-organization and differentiation in stem cell and organoid models
Miguel Silva

P070 Investigating the role of LZTR1 in noonan syndrome-associated brain development using iPSC-derived models
Ana Trpchevska

P071 Immune checkpoint molecules, the secrets of stem cell privilege in the lung
Roxana Maria Wasnick

P072 iPSC-derived choroid plexus organoids for modeling Huntington's disease
Wenqing Xu

Abstract No. P052

Automated high-throughput generation and characterization of human neuromuscular organoids

Ines Lahmann[1], *Inês Afonso Martins*[1,*], *Ina-Maria Rudolph*[2], *Anna R. Hashtchin*[1], *Sebastian Diecke*[2], and *Mina Gouti*[1]

[1] Stem cell modeling and disease lab, Max Delbrück Center for Molecular Medicine, Berlin
[2] Pluripotent stem cell technology platform, Max Delbrück Center for Molecular Medicine, Berlin

*Presenting author

Human neuromuscular organoids (hNMOs) represent a complex and physiologically relevant system that provides unprecedented opportunities for various applications within the neuromuscular field, ranging from developmental studies to disease modelling and drug testing. However, traditional manual workflows for organoid research are labour-intensive, time-consuming, difficult to scale, and highly susceptible to user variability and human error.

To address these challenges, we are developing and implementing a fully automated high-throughput workflow for hNMO research, encompassing generation, imaging, and analysis. For the generation of hNMOs, we employed a customised liquid-handling robotic system programmed to replicate the necessary steps, followed by spinner-flask culture to facilitate large-scale production. Comparative analyses revealed greater consistency in size at early stages, as well as morphological and molecular similarities between manually and robotically derived organoids. For organoid characterisation, we used a High-Content Screening System for image acquisition and developed a customized machine learning-based pipeline for subsequent image analysis. Results from neuromuscular junction and innervation assessments demonstrate that the automated tool can capture trends between conditions, which were previously observed through manual analysis using Fiji.

The automated high-throughput generation and characterization of hNMOs have the potential to revolutionize the field of organoid research. This workflow promises to significantly accelerate disease modelling and drug discovery.

Abstract No. P053

Label-free functional analysis for the screening of iPSC-derived neural organoid response to neuroactive compounds

Ben Streeter, Timm Schlegel, Denise Sullivan, Austin Passaro, Stacie Chvatal, Daniel Millard*

Axion BioSystems, Eindhoven, The Netherlands

*Presenting author

In vitro neuronal models are valuable tools for studying brain function, neural diseases, and toxicology. Recent advances in induced pluripotent stem cell (iPSC) technology have allowed researchers to produce 3D models of neural tissue, termed neural organoids, that better recapitulate the cellular diversity and spatial architecture of *in vivo* tissue. Here, a workflow to monitor the differentiation of neural organoids and their response to neuroactive compounds is described. Neural organoids were both generated via commercially available kits and purchased as a finished product for this study. To generate neural organoids, iPSCs were first plated and monitored using a whole-vessel imaging system (the Omni). Colony area, diameter, and coverage increased over a three-day period, at which point the colonies reached an optimal size for passaging and subsequent embryoid body formation. Following neural induction using STEMCELL Technologies' Dorsal Forebrain Organoid Differentiation Kit, organoids in suspension culture were imaged for 50+ days using the Omni, and organoid size and count were monitored, illustrating the progression of the differentiation process. For electrophysiological measurements, pre-formed Human iPSC-Derived Midbrain Organoids from STEMCELL Technologies were cultured using the STEMdiff™ Neural Organoid Maintenance Kit. The midbrain organoids were plated onto CytoView 6 MEA plates and, on Day 125 post-differentiation, dosed with neuroactive compounds or a DMSO control. Baseline and post-dose (1 hour after drug addition) recordings were taken using the Maestro Pro and showed robust changes in midbrain organoid activity patterns in response to 4-aminopyrirdine and rotenone. Dosing with 4-aminopyrirdine increased the mean firing rate and network burst frequency of midbrain organoids, while dosing with rotenone led to a decrease in both metrics. In total, this study shows that the Omni and Maestro Pro systems are valuable tools for studying the differentiation of neural organoids and their subsequent use in toxicological screening applications.

Abstract No. P054

Role of dermal-epidermal junction topography in regulating epidermal stem cell fate

Adil Boolani[*], and *Fiona Watt*

EMBL, Heidelberg

*Presenting author

Our skin is divided into two layers - the epidermis and the dermis. The epidermis overlays the dermis and the interface between these two layers, called the dermal-epidermal junction (DEJ), undulates forming the rete ridges. The rete ridges do not remain the same throughout our lives but flatten out as we age. The cells of the epidermis, known as keratinocytes, are connected to each other by cell-cell junctions and to the extracellular matrix (ECM) by cell-ECM junctions. In healthy skin, epidermal stem cells, which divide and differentiate to maintain the epidermis, lie between the rete ridges (Jones et al, 1995). Though epidermal stem cells are known to have a non-random distribution in the skin, it is not clear how this patterning is controlled and whether the change in topography of the rete ridges during aging contributes to the concurrent loss in expression of stem cell markers.

To dissect the underlying mechanisms controlling epidermal stem cell patterning, I am using a previously developed *in vitro* model system based on polydimethylsiloxane substrates that recapitulates stem cell patterning and the dimensions of the DEJ of healthy (undulating substrates) and aged skin (flat substrates)(Mobasseri et al, 2019). Using a combination of molecular tension sensors, fluorescence lifetime imaging microcopy and immunostaining, I find that cells located at different positions on the undulating substrates experience different magnitudes of tension at cell-cell and cell-ECM junctions. Moreover, cell multilayers experience homogeneous levels of tension at cell-ECM junctions when grown on flat substrates compared to undulating substrates. This suggests that epidermal stem cell identity, and, in turn, patterning, may be regulated by the topography of the DEJ.

Jones PH, Harper S & Watt FM (1995) Stem cell patterning and fate in human epidermis. Cell 80: 83–93

Mobasseri SA, Zijl S, Salameti V, Walko G, Stannard A, Garcia-Manyes S & Watt FM (2019) Patterning of human epidermal stem cells on undulating elastomer substrates reflects differences in cell stiffness. Acta Biomaterialia 87: 256–264

Abstract No. P055

Enhancing consistency in 3D spheroid engineering: A novel automated cell picking approach

Daryl Cole, and *Nicola Bevan*

Sartorius AG, Royston, UK

Presenting author – see below

Creating 3D models for drug development and disease modeling presents several challenges. Traditional methods for generating these models are often slow, resource-intensive, and yield inconsistent results. Automated technologies provide solutions by streamlining complex processes, resulting in time and resource savings while enhancing workflow robustness and consistency. The CellCelector Flex, an automated cell picking platform, addresses the limitations of conventional methods and, when used in conjunction with other bioanalytical instruments, supports a simplified workflow.

In this study, spheroids were cultured from individual Nuclight green-expressing iPSCs to produce spheroids from a single cell, resulting in monoclonal 3D spheroid structures. Engineering these monoclonal iPSC spheroids allows for the customization of models to specific diseases or conditions and offers insights into 3D structure development using fluorescently labeled cell populations.

iPSCs were grown in S200 nanowell plates, and spheroid formation was monitored using the Incucyte® Live-Cell Analysis System and CellCelector over three days. Wells containing growing spheroids originating from a single cell were identified for picking. Each spheroid, from its individual nanowell, was picked, resuspended in hydrogel, and embedded into a well of medium in a 96-well ULA plate. Spheroids exhibited steady proliferation over the next six days, expanding 14-fold. Post-dissociation, cells were assessed for viability and expression of SSEA-4 and TRA-1-60, markers of pluripotency, using the iQue® HTS Cytometer. Cells within the spheroid showed a comparable phenotype to the original iPSC cells (>95% viability, >95% pluripotency). Additionally, a similar workflow was employed for selecting and culturing iPSC-derived hepatic organoids directly from Matrigel domes, demonstrating retained growth and phenotype characteristics.

In conclusion, this approach simplifies the culture and generation of organoids and monoclonal spheroids from iPSCs for drug discovery, development, and toxicity studies. Future developments of this workflow aim to generate organoids from a single iPSC using the techniques described in this poster.

This poster presentation will be presented by Sofia Kuzmich (Sartorius).

Abstract No. P056

Cellular plasticity and replenishment capacity in human brain organoids

Beatriz Lopez-Amo Calvo, *Lara Harringer*, *Frank Edenhofer*, and *Christopher Esk*[*]

Institute of Molecular Biology, University of Innsbruck, Austria

*Presenting author

Brain development comprises neural stem cells that expand by symmetric cell division before switching to asymmetric cell divisions maintaining neural stem cells while producing differentiating functional cell types of the brain. The timing of neural stem cells' shift between these division modes varies between species and individual cells thereby generating heterogeneity in lineages' sizes and contribution to the developing brain. Little is known about tissue-level control of these processes. Experiments in human brain brain organoids using whole tissue lineage tracing, computational modelling and lineage-resolved scRNA-seq have demonstrated how neural progenitor cells follow a dynamic growth pattern. Progenitor derived lineages vary in size with large lineages maintaining a pool of symmetrically dividing cells well into neurogenesis. Experiments in chimeric organoids containing control and growth perturbed cells revealed that wildtype cells compensate for growth defects in perturbed lineages. This indicates that neural stem cell fate is plastic and that mechanisms of tissue-mediated growth control exist in the developing human brain. Partial ablation experiments in chimeric organoids have been performed to characterize the replenishment capacity of long-lasting neural stem cells at different stages of organoid development and neurogenesis. Microscopic and flow cytometry analyses reveal variable phenotypic rescue 4 weeks after cell perturbation depending on ablation timing. Ablated organoids maintain similar ratios of progenitors to neurons compared to untreated brain organoids during tissue replenishment. Cell cycle analyses indicate changes in cell cycle dynamics among replenishing cells. These results, together with transcriptional analysis and lineage-resolved scRNA-seq characterize cellular plasticity and replenishment capacity of developing human brain tissue.

Abstract No. P057

High fat diet affects the stromal niche signaling in the porcine intestine

Falk Janos Farkas[1,], Michael Sterr[2], Tobias Greisle[1], Mireia Molina van den Bosch[1], Simone Renner[3], Christos Karampelias[2], Juliane Merl-Pham[4], Christine von Törne[4], Stephanie Hauck[4], Eckhard Wolf[3], Anika Böttcher[1]*, and *Heiko Lickert[1]*

[1] Institute of Diabetes and Regeneration, Helmholtz Munich

[2] Helmholtz Diabetes Center, Helmholtz Munich

[3] Center for Innovative Medical Models, Ludwig-Maximilians-University Munich

[4] Research Unit Protein Science Helmholtz Munich

*Presenting author

Diet-induced obesity is a global health burden that predisposes to type 2 diabetes and colorectal cancer. The intestine, as the body's largest endocrine organ, plays a key role in regulating glucose homeostasis and energy balance. Obesogenic diets in mice disrupt intestinal homeostasis, inducing stem cell hyperproliferation, altered differentiation, shifts in enterocyte regional identity, and enteroendocrine reprogramming. While rodent studies implicate both epithelial and subepithelial compartments in governing the intestinal stem cell (ISC) niche, the impact of a high-fat diet (HFD) on this niche remains unclear. Our initial co-culture experiments using mouse intestinal organoids and HFD-derived mesenchymal niche showed enhanced organoid formation, indicating increased niche-derived proliferative cues in HFD. However closer characterization of the subepithelial niche – especially in large animal models – is still lacking.

To address this, we employed the pig as a physiologically relevant model and analyzed small and large intestinal tissue following a 30-week HFD intervention, including both calorie-restricted and obesogenic HFD groups. Using a novel microdissection technique, we isolated ISCs together with their surrounding subepithelial niche. We then conducted integrative histological, proteomic, and single-nucleus transcriptomic profiling to assess how dietary fat alters niche signaling and epithelial differentiation.

Our data reveal reduced levels of proteins involved in metabolism and membrane transport, alongside histological signs of elevated proliferation and loss of specific enteroendocrine populations. Proteomic and transcriptomic data both show significant remodeling of the subepithelial mesenchyme in HFD-fed animals, including upregulation of VEGF, PDGF, and EGF signaling pathways, along with changes in extracellular matrix composition.

These findings indicate that dietary fat disrupts the ISC niche and impairs epithelial differentiation, independent of caloric intake. This work provides a foundational multi-modal dataset from a large animal model, offering new insight into how HFD drives early intestinal maladaptation underlying metabolic dysfunction and increased colorectal cancer risk.

Aliluev A, Tritschler S, Sterr M, [...] Lickert H, Böttcher A. Diet-induced alteration of intestinal stem cell function underlies obesity and prediabetes in mice. Nat Metab. 2021 Sep;3(9):1202-1216. PMID: 34552271

Enriquez JR, McCauley HA, Zhang KX, Sanchez JG, Kalin GT, Lang RA, Wells JM. A dietary change to a high-fat diet initiates a rapid adaptation of the intestine. Cell Rep. 2022 Nov 15;41(7):111641. doi: 10.1016/j.celrep.2022.111641. PMID: 36384107

Abstract No. P058

TBR2 directs neuronal subunits switching in ATP-dependent chromatin remodeling complexes to regulate chromatin accessibility during human cortical development

Catello Guida[1,2,*], *Matteo Gasparotto*[1,3], *Anne Hoffrichter*[1,3], *Fabio Marsoner*[1,3], *Annasara Artioli*[1,2], and *Julia Ladewig*[1,3]

[1] Zentralinstitut für Seelische Gesundheit, Mannheim
[2] Heidelberg University
[3] German Cancer Research Center (DKFZ), Heidelberg

*Presenting author

The human neocortex develops through a complex interplay of metabolic, transcriptional, and cellular processes. A key transition in neurogenesis involves the shift from aerobic glycolysis, used by neural progenitor cells (NPCs), to oxidative phosphorylation (OXPHOS), supporting both neural differentiation and chromatin remodeling. This works in concert with a tightly regulated transcriptional cascade – PAX6, TBR2, and TBR1 – that directs progenitor proliferation, neuronal specification and cortical layer formation. Disruptions in these processes have been linked to severe neurodevelopmental disorders. We investigated the role of TBR2 in human cortical development through a CRISPR/Cas9-mediated TBR2 knockout (KO) in human induced pluripotent stem cells, and derived cortical organoids. Loss of TBR2 impaired NPC differentiation and disrupted neuronal subpopulation emergence, accompanied by altered mitochondrial morphology in progenitors and neurons. Single-nuclei transcriptomic and ATAC-seq data revealed a downregulation of ribosomal and OXPHOS pathways and significant chromatin accessibility changes in neuronal populations. This enabled the identification of NuRD and nBAF chromatin remodeling complex components as novel targets of TBR2. These findings highlight TBR2's crucial role in integrating metabolic, transcriptional, and epigenetic pathways during cortical development, underscoring the power of human organoid models to elucidate mechanisms underlying neurodevelopmental disorders.

Xinde Zheng,Leah Boyer,Mingji Jin,Jerome Mertens,Yongsung Kim,Li MaLi Ma,Michael Hamm,Fred H Gage,Tony Hunter (2016) Metabolic reprogramming during neuronal differentiation from aerobic glycolysis to neuronal oxidative phosphorylation eLife 5:e13374.

Andrew S Yoo, Gerald R Crabtree, ATP-dependent chromatin remodeling in neural development, Current Opinion in Neurobiology, Volume 19, Issue 2, 2009, Pages 120-126, ISSN 0959-4388,

Abstract No. P059

Scalable, defined cardiac organoids for drug discovery and non-cryogenic distribution

Sarkawt Hamad[1,*], *Ebru Aksoy*[1,2], *Maria Kalil*[3], *Daina Martínez-Falguera*[3], *Rezwan Firuzi*[1], *Albert Teis*[4], *Julia Aranyó*[5], *Esther Jorge*[3], *Felipe Bisbal*[4], *Antoni Bayés-Genís*[3], *Carolina Gálvez-Montón*[3], and *Kurt Pfannkuche*[1]

[1] Institute for Neurophysiology, University of Cologne
[2] Marga-and-Walter-Boll Laboratory for Cardiac Tissue Engineering, University of Cologne
[3] Research Institute Germans Trias i Pujol, Badalona, Spain
[4] Carlos III Health Research Institute, Badalona, Spain
[5] Autonomous University of Barcelona, Spain

*Presenting author

Cardiac organoids (COs) derived from human induced pluripotent stem cells (hiPSCs) are emerging as powerful tools for disease modeling and drug discovery, yet scalable and defined culture systems remain elusive. We developed a robust, serum- and matrix-free, chemically defined platform for generating self-organizing COs in 96-well plates and scalable bioreactor suspension systems. These COs comprise cardiomyocytes (CMs), cardiac fibroblasts (cFBs), endothelial cells (cECs), smooth muscle cells (cSMCs), pericytes, and other cardiac-relevant populations.

We characterized COs via immunostaining, transmission electron microscopy, flow cytometry, and single-cell RNA sequencing. Within 20 days, the COs had developed organised sarcomeres (approximately 1.7 - 2 µm in length), expressed cardiac maturation markers (e.g. RYR2 and PLN) and contained a variety of cardiac cell types, including CMs, cFBs, cECs, cSMCs, and proliferative non-myocytes such as pericytes. To test the feasibility of lab-to-lab distribution, approximately 7,000 COs were shipped internationally under non-cryogenic conditions (without quality loss). Following a 24-48 hours recovery period, electrophysiological analysis using high-density microelectrode arrays (HD-MEAs) revealed that the COs had retained their electrophysiological function.

Under control conditions, the COs exhibited synchronised contraction at a rate of 54.09 ± 2.35 cardiac field potentials (cfp) / min. Adrenergic stimulation increased beat rate and signal amplitude (96.46 ± 2.26 cfp/min), whereas KCl maintained high frequency (98.80 ± 1.77 cfp/min) but disrupted conduction patterns, highlighting pharmacodynamic sensitivity.

These results demonstrate the generation of functionally mature-like cardiac organoids in a defined, scalable system, suitable for drug discovery and non-cryogenic distribution. This platform enables reliable pharmacological screening and holds promise for future regenerative cardiac therapies.

Key words: cardiac organoids, bioreactor, scalable, drug discovery

Abstract No. P060

Pancreatic ductal organoids for basic developmental biology and drug screening applications

Christos Karampelias[1,*], *Kaiyuan Yang*[1], *Falk Janos Farkas*[2], *Michael Sterr*[1], *Mireia Molina van den Bosch*[2], *Simone Renner*[3], *Janina Fuss*[5], *Sören Franzenburg*[5], *Christine von Törne*[5], *Tatsuya Kin*[6], *Eckhard Wolf*[3], *Elisabeth Kemter*[3], and *Heiko Lickert*[2]

[1] Helmholtz Diabetes Center, Munich
[2] Institute of Diabetes and Regeneration Helmholtz Munich
[3] Center for Innovative Medical Models (CiMM), Ludwig-Maximilians-University, Munich
[4] Institute of Clinical Molecular Biology (IKMB), UKSH Kiel
[5] Research Unit Protein Science, Helmholtz Munich
[6] Clinical Islet Laboratory, University of Alberta, Canada

*Presenting author

The existence of adult pancreatic progenitors is a long-debated topic in the pancreas field. Given the difficulty to access pancreas tissue from patients and the scarcity of donor material, new and well characterized tools are needed to study pancreatic progenitor populations and develop new regenerative therapies. Pancreatic organoids derived from exocrine tissue are a great innovation to study basic progenitor biology, disease mechanisms and screen for new therapeutics in vitro and in a high-throughput manner. In this work, we characterized porcine pancreatic ductal organoids as an alternative model for high-throughput pancreatic drug screening. Pigs share great similarities in terms of size and metabolism to the human system. We performed an in-depth benchmarking of porcine pancreatic ductal organoids using single cell RNA sequencing and proteomics against human-derived pancreatic organoids and primary pancreatic tissue and identified the similarities between the systems. We further assessed the range of applications that these porcine pancreatic organoids can be applied for, by assaying organoids derived from distinct developmental stages. Lastly, we used porcine pancreatic ductal organoids to screen FDA-approved drugs as inducers for drug-induced pancreatitis and identified causative agents that can induce pancreatic cell death. We translated the most promising primary candidates to the human pancreatic organoid system highlighting the usefulness of this model for translational applications. Our work paves the way for use of an easily obtainable pancreatic exocrine source (porcine ductal organoids) for developmental biology and drug screening applications with comparable results to the hard to obtain human pancreatic tissue.

Abstract No. P061

Modeling radiation-induced neurotoxicity in human cerebral organoids

Leon Kaysan[*], *Tamara Bender*, and *Insa Schröder*

GSI Helmholtzzentrum für Schwerionenforschung GmbH, Darmstadt

*Presenting author

Radiotherapy is critical in treating brain tumors, yet it carries the risk of damaging adjacent healthy tissue, with long-term neurocognitive consequences. While rodent models have highlighted radiation-induced synaptic loss and altered neuronal function translating these findings to the human brain remains challenging. Here, we employ human cerebral organoids to develop a human-specific model of radiation-induced neurotoxicity.

We apply clinically relevant regimens of X-ray and carbon heavy ion irradiation to mature organoids and assess the effects on neuronal networks using a combination of structural and functional readouts. Synaptic integrity is evaluated by quantitative imaging, and neuronal activity is monitored using jGCaMP8-based calcium imaging. These parallel assessments aim to capture both anatomical disruption and real-time functional changes post-irradiation.

Our approach builds on prior brain tumor-organoid co-culture experiments, where these same radiation doses effectively reduced tumor viability. By turning attention to non-tumor organoids, we seek to understand how therapeutic irradiation impacts healthy neural tissue. This model system provides a novel platform for investigating the cellular basis of radiation-induced cognitive decline and may contribute to optimizing radiotherapy strategies in neuro-oncology. It is expected to be applied to other organoid models e.g. to better understand the radiosensitivity of certain brain regions like the hippocampus and prefrontal cortex.

Schickel, E. et al. Human cerebral organoids model tumor infiltration and migration supported by astrocytes in an autologous setting. 2025.01.29.635456 Preprint at https://doi.org/10.1101/2025.01.29.635456 (2025).

Abstract No. P062

Human otic Bioengineered Neuronal Organoids (oBENOs) mimic early inner ear development and physiology

Angeliki Koufali[1,*], *Anika K. Müller*[2], *Norbert Babai*[1], *Bao Guobin*[1], *Aleyna M. Diniz*[1], *Emre Taylan Duman*[1], *Eunseo Kim*[1], *Jette Alfken*[3], *Michael Setya*[1], *Kea Schmoll*[1], *Tim Salditt*[3], *Kathrin Kusch*[1], *Tobias Moser*[1], and *Maria-Patapia Zafeiriou*[1]

[1] University Medical Center, Georg-August-University, Göttingen
[2] University of Copenhagen, Copenhagen, Denmark
[3] University of Göttingen

*Presenting author

Hearing impairment, the most common sensory disorder, affects millions of people worldwide. The most prevalent form, sensorineural hearing loss (SNHL), is traced back to the loss of hair cells (HCs), the mechanosensory cells responsible for detecting sound and movement, which lack regenerative capacity in vertebrates. HC loss can, in turn, lead to the degeneration of spiral ganglion neurons (SGNs), which transmit auditory signals to the auditory cortex. Understanding the mechanisms of SNHL and developing effective treatments remain critical to addressing this major unmet medical need. Otic organoid models offer a promising platform for disease modeling, drug screening, and regenerative medicine. Here, we report the generation of a robust human induced pluripotent stem cell (hiPSC)-derived organoid model, termed the otic Bioengineered Neuronal Organoid (oBENO), which reproducibly develops both functional HCs and SGNs, along with ribbon synapses. Temporal bulk RNA sequencing analysis revealed that oBENO development closely recapitulates *in vivo* early inner ear formation, marked by the presence of several inner ear-specific cell populations alongside an intricate vascular network. Retinoic acid addition enhanced HC and SGN maturation while supressing the development of non-otic lineages such as epidermal cells. Furthermore, we generated a transgenic reporter line (trSGN) to enable live-cell imaging and functional analysis of Parvalbumin-positive SGNs. Patch-clamp and calcium imaging characterized the function of HCs and SNGs, revealing that endogenous glutamate neurotransmission drives SGN activation in oBENO, thereby modeling physiology. The defined multicellular composition, reproducibility, and functional properties of oBENOs render them promising models for studying inner ear development and for evaluating therapies towards sensory hearing loss restoration.

Abstract No. P063

Modulation of ERK signaling dynamics by pharmacological intervention directs cell differentiation

*Pavlina Markova**, *Purna Gadre*, *Joanna Sobska*, and *Fiona Watt*

EMBL, Heidelberg

*Presenting author

The extracellular signal-regulated kinase (ERK), a central component of the Ras-Raf-MEK-MAPK signaling pathway, orchestrates critical cellular processes including proliferation, differentiation, and survival. Emerging evidence highlights that ERK dynamics play a pivotal role in regulating gene expression and influencing developmental processes, adult tissue homeostasis, and wound healing. However, how ERK signaling patterns translate into binary cell states remains poorly understood.

Recent work by Hiratsuka et al. (2020) revealed that individual keratinocytes transition through distinct ERK activity states—from high, sustained ERK activity associated with the stem cell state, to pulsatile activity, and then to low, sustained activity characteristic of differentiation. ERK pulses are induced by differentiation-promoting cues, such as reduced β1 integrin-mediated adhesion and increased cell density. Despite these insights, the functional significance of pulsatile ERK activity in controlling cell fate transitions is still unclear. Here, we combine live-cell imaging of keratinocyte colonies transfected with a real-time ERK kinase translocation receptor biosensor and pharmacological modulation of ERK dynamics to examine the role of pulses in regulating epidermal homeostasis.

We observe that inhibition of dynamin-mediated endocytosis using Dyngo-4a reduces overall ERK activity, but increases the number of pulsing cells. Furthermore, we noted that Dyngo-4a treatment promotes keratinocyte differentiation. To elucidate the mechanism underlying this observation, we investigated whether reduction in ERK activity influences Notch signaling, a positive regulator of keratinocyte differentiation. We found that reduction in ERK activity by MEK inhibition reduces the expression of a Notch ligand, Jagged1. We hypothesize that pulsatile ERK activity induces heterogeneity in Jagged1 levels within keratinocyte neighbours to induce symmetry breaking events that may drive differentiation. Our work provides novel insights into the spatiotemporal control of the onset of differentiation.

Hiratsuka T, et al. Proc Natl Acad Sci U S A. 2020 Jul 28;117(30):17796-17807. doi: 10.1073/pnas.2006965117.

Goglia AG, et al. Cell Syst. 2020 Mar 25;10(3):240-253.e6. doi: 10.1016/j.cels.2020.02.005.

Abstract No. P064

Organoid assembloids reveal serotonergic regulation of human cortical development

Raquel Pérez Fernández[1,*], *Marco T. Siekmann*[1], *Martin Kubitschke*[2], *Lutz Wallhorn*[2], *Ammar Jabali*[1], *Anne Hoffrichter*[1], *Annasara Artioli*[1], *Matteo Gasparotto*[1], *Philipp Koch*[1], *Olivia Andrea Masseck*[2], and *Julia Ladewig*[1]

[1] Central Institute of Mental Health, Mannheim
[2] University of Bremen

*Presenting author

The orchestration of human cortical development requires precise coordination of cellular processes, with serotonin (5-HT) as a key modulator. Here, we developed an in vitro system combining induced pluripotent stem cell (iPSC)-derived neural progenitors, cortical organoids (COs), and raphe organoids (ROs) to investigate serotonergic regulation of cortical development.

Exposure of cortical progenitors and COs to 5-HT enhanced proliferation in apical (aRG) and basal radial glial cells (bRG) through distinct receptor subtypes: 5-HTR2C mediates aRG proliferation, while 5-HTR2A controls bRG proliferation. To establish a physiological 5-HT source, we generated ROs containing ventral hindbrain progenitors ($NKX2.2^+$/$FOXA2^+$) that differentiated into serotonergic neurons expressing TPH2, VMAT2, and 5-HT. Single-nuclei RNA sequencing of ROs revealed these serotonergic progenitor and neuronal populations. Extending this approach, we also profiled COs and cortical–raphe assembloids at single-nucleus resolution, to identify region-specific cell types and transcriptomic changes in response to serotonergic innervation. Functional validation using the sDarken biosensor confirmed the ROs' capacity to release and bind 5-HT.

We then established cortical–raphe assembloids that reproduced physiological innervation patterns of developing cortex by serotonergic projections. These assembloids exhibited extensive serotonergic innervation throughout the cortical tissue, with enhanced progenitor proliferation specifically in regions receiving 5-HT projections.

These findings demonstrate that serotonin promotes cortical development through selective enhancement of progenitor proliferation via specific receptor subtypes. By combining functional and transcriptomic profiling, this organoid-based platform provides a powerful model system for investigating serotonergic regulation of human corticogenesis and its potential dysregulation in neurodevelopmental disorders.

Abstract No. P065

CSF1R-dependent myeloid cells direct hepatocyte development in human and mouse models

Susanna Quach[1,*], *Viktor Glaser*[2], *Julian Weihs*[1], *Yeni Ait Ahmed*[1], *Adrien Guillot*[1], *Kerim Acil*[3], *Elvira Mass*[3], and *Milad Rezvani*[1]

[1] Charité Universitätsmedizin Berlin
[2] Berlin Institute of Health (BIH) at Charité, Berlin
[3] Life and Medical Sciences (LIMES) Institute, University of Bonn

*Presenting author

The embryonic liver serves as a developmental niche, facilitating the differentiation of bipotent hepatoblasts into hepatocytes or biliary cells. Liver-resident macrophages, the most abundant myeloid population in the liver, co-develop from yolk sac–derived hematopoietic progenitors. While myeloid-to-hepatobiliary crosstalk is well-studied in pathology, its role in hepatic versus biliary fate decision during embryonic development remains elusive.

We utilized multi-tissue fetal liver organoids (FLOs), derived from human induced pluripotent stem cells. FLOs establish an autonomous niche supporting the differentiation of HepPar1+ AFP+ hepatocyte-like cells, CK19+ biliary-like cells, and IBA-1+ myeloid-like cells with yolk sac ontogeny.

We investigated the role of myeloid cells in hepatobiliary development in different models.

a) *In vitro*, we depleted CD45+ CD14+ myeloid cells in FLOs using both chemical CSF1R inhibition and CRISPR-mediated *CSF1R* knockout.

b) *In vivo*, we depleted yolk sac–derived macrophages from embryonic day (E) 12.5 and E14.5 onwards using conditional *Csf1r* and *Spi1* knockouts, respectively.

In vitro, depletion of CD45+ CD14+ cells reduced the ALB+ PanCK- hepatocyte-like population ($p = 0.0015$), but not proliferation, while preserving the bipotent ALB+ PanCK+ progenitor pool. Further, myeloid depletion downregulated expression of the hepatocyte transcription factor and myeloid activation regulator *CCAAT/enhancer binding protein beta* (*C/EBPβ*, $p = 0.039$) and the functional hepatocyte marker *haptoglobin* (*HP*, $p = 0.0003$). *In vivo*, early macrophage depletion from E12.5 significantly reduced hepatocyte nuclear factor 4 alpha expression (Hnf4α, $p = 0.0306$), whereas depletion from E14.5 had no significant effect ($p = 0.0904$), indicating that macrophages promote hepatocyte fate during a regulated developmental window.

In summary, human organoid and mouse models support a role for CSF1R-dependent myeloid cells in promoting hepatocyte fate from a bipotent progenitor pool. Future work, including single-cell RNA sequencing and functional validation in FLOs, will further define mechanisms of myeloid–hepatic crosstalk.

Abstract No. P066

Self-organizing vascularization in human neuromuscular organoids drives functional maturation and regenerative capacity

*Anna Rafiei Hashtchin**, *Caroline Beyer*, *Sara De Vincentiis*, *Sandra Cristina Carneiro Raimundo*, *Chrysanthi-Maria Moysidou*, *Noelle Findeisen*, *Iacopo Bicci*, *Anje Sporbert*, and *Mina Gouti*

Max-Delbrück-Center for Molecular Medicine, Berlin

*Presenting author

Vascularization remains a major hurdle in improving the viability, maturation, and functional relevance of organoid systems. Here, we present a developmentally guided strategy for intrinsic vascularization of human neuromuscular organoids (vNMOs), leveraging the self-organizing capacity of neuromesodermal progenitors. Through temporal modulation of extracellular signals, we induce transient expression of key endothelial regulators, leading to the emergence of endothelial cells that organize into extensive, spatially patterned vascular networks throughout the organoids.

These vascular structures are supported by mural cells expressing smooth muscle actin and pericyte markers, and 3D lightsheet imaging confirms the formation of interconnected vessel-like networks. Importantly, the vascular niche develops in parallel with neural and muscle compartments without disrupting their organization. vNMOs also support the spontaneous appearance of microglia-like immune cells. Enhanced neuromuscular junction (NMJ) formation is observed, characterized by increased NMJ number and size, alongside retained contractile function. Upon mechanical injury, vNMOs initiate a regenerative response involving satellite cell activation and immune cell recruitment.

This approach eliminates the need for exogenous endothelial co-culture, offering a scalable and robust method for intrinsic vascularization. Our findings highlight the potential of spatiotemporal signaling modulation to drive vascular lineage specification within organoids, enabling the formation of self-organized vascular systems that support tissue development and repair. vNMOs serve as a powerful platform to study neuromuscular development, neurovascular interactions, and regenerative mechanisms, with broad applications in disease modeling and therapeutic discovery.

Abstract No. P067

Exploring the regional identity and chromatin landscape of human embryonic neural stem cells

Amelie Schurer[1,], Angeliki Spathopoulou[1], Marcel Tisch[1], Katharina Günther[1], Marc-Christian Thier[2], Marta Suarez Cubero[1], Theresa Lindlbauer[1], Philipp Wörsdörfer[3], Erhard Wischmeyer[4], Chee Keong Kwok[3], Louisa Dury[1], Antonella Karin Rokita[1], Li Li[5], Julia Schlee[6], Julia Flunkert[7], Thomas Haaf[7], Tanja Schlaiß[8], Simone Liebscher[9], Nicole Wagner[3], Francesca Finotello[1,10], Süleyman Ergün[3], Gunnar Schotta[5], Christopher Esk[1], Oliver Brüstle[6], Katja Schenke-Layland[9,11], Andreas Trumpp[2]*, and *Frank Edenhofer[1]*

[1] Institute of Molecular Biology, Department Genomics, Stem Cell Biology & Regenerative Medicine, Leopold-Franzens-University Innsbruck, Austria; **[2]** German Cancer Research Center - HI-STEM, Heidelberg; **[3]** Inst. Anatomy & Cell Biology, Stem Cell and Regenerative Medicine Group, Uni. Würzburg; **[4]** Medical Faculty OWL, Cellular Neurophysiology, Uni. Bielefeld; **[5]** Biomedical Centre (BMC), Ludwig-Maximilians-University, Munich; **[6]** Institute of Reconstructive Neurobiology, University Hospital Bonn; **[7]** Institute of Human Genetics, Biocenter, University of Würzburg; **[8]** Department of Gynecology and Obstetrics, University Hospital Würzburg; **[9]** Inst. Biomed. Engineering, Dep. Med. Tech. & Reg. Med., Eberhard Karls Uni. Tübingen; **[10]** Digital Science Center (DiSC), University of Innsbruck; **[11]** NMI Natural and Medical Sciences Institute at the University Tübingen, Reutlingen

*Presenting author

A complete understanding of the earliest human specific neurodevelopmental processes remains a significant challenge in the field of developmental biology. In the past, primarily animal models were employed to model these processes, however in the context of human neurodevelopment they lack complexity and similarities to the human organism. Previous studies described rosette-like progenitors and PSC-derived neuroepithelial cells (Cattaneo and Conti, 2010), yet the establishment of a stable, tissue-derived human neural progenitor cell (NPC) line has remained elusive. Previously, we established a long-term *in vitro* maintained embryonic neuroepithelial stem/progenitor cell (eNSPC) line, stabilized in chemically defined medium. These cells exhibit a naïve, non-polarized phenotype and as confirmed by single-cell RNA sequencing, represent a homogeneous population of multipotent stem cells. eNSPCs demonstrate a broad differentiation potential, generating forebrain, midbrain, and hindbrain identities and neural crest lineages, highlighting their capacity to produce diverse central and peripheral nervous system derivatives. Here, we present the analysis of their regional identity along the anterior-posterior and dorsal-ventral axes, leading us to hypothesize that eNSPCs originate from the midbrain-hindbrain border (MHB) region. Moreover, we found changes in the eNSPCs chromatin accessibility profile over prolonged culturing and a distinct epigenetic landscape compared to other NPC populations. Principal component analysis revealed distinct clustering of eNSPCs separated from induced neural stem cells (iNSCs) and small molecule NPCs (smNPCs). On a single-locus level, high-passage eNSPCs exhibited increased chromatin accessibility at the regulatory region of the *SOX2*, while accessibility of neurogenic markers such as *ASCL1* remained unchanged, suggesting a reinforced stem cell identity and stabilization of an early multipotent neural progenitor state upon prolonged culture in CSPFL medium. Taken together, the establishment and comprehensive characterization of eNSPCs provides a robust platform to study human specific processes in early neurodevelopment and potentially serves as a novel tool for disease modelling and advancing regenerative therapies.

Conti, L., Cattaneo, E. Neural stem cell systems: physiological players or in vitro entities?. Nat Rev Neurosci 11, 176–187 (2010). https://doi.org/10.1038/nrn2761

Abstract No. P068

Reproducible and efficient organoid development using stable and highly active synthetic peptide alternatives to BDNF, Noggin, and Wnt3a

Bärbel Icheln[1], *Kazuki Shigematsu*[2,*], and *Kosuke Minamihata*[2]

[1] Active Bioscience GmbH, Hamburg
[2] PeptiGrowth, Tokyo, Japan

*Presenting author

Generation of neural, pulmonary, and definitive endoderm organoids requires reliable and consistent growth factor signaling, which conventional recombinant GFs often fail to provide due to variability, impurities, instability, and high costs. To address these issues, we have developed synthetic peptides that replicate the functions of conventional GFs while overcoming these limitations.

This poster highlights three synthetic peptide growth factors designed to enhance these organoid systems:

• Neural organoids: Using Nerve Organoids, a proprietary system developed by Jiksak Bioengineering, PG-003 enhanced neural axon growth, producing longer and thicker axon bundles compared to rBDNF.

• Pulmonary organoids: Noggin Like Peptide, PG-004 effectively induced the differentiation of lung progenitor cells from hiPSCs and facilitated the formation of lung alveolar organoids at levels comparable to or succeeding those achieved with Noggin.

• Definitive endoderm organoids: Using WNT3a alternative Peptide, PG-008, Wnt/β-catenin signaling was activated, achieving efficient differentiation from iPSCs. While CHIR99021 often faces issues of cytotoxicity, PG-008 exhibits no cytotoxicity up to 10 µM, the highest concentration tested. This contributes to increased development efficiency for various types of organoids without the cytotoxicity observed with CHIR99021.

These synthetic peptides provide stable, reproducible, and defined tools for organoid research, facilitating standardized and cost-effective generation of complex tissue models.

Abstract No. P069

Label-free live cell imaging for spatiotemporal quantification of self-organization and differentiation in stem cell and organoid models

Miguel Silva[1,*], *Anna Jötten*[2], and *Philipp Paulitschke*[3]

[1] Technical University of Munich (TUM)
[2] Ludwig Maximilian University of Munich (LMU)
[3] Phio Scientific, Munich

*Presenting author

Longitudinal, label-free imaging of live cells within physiologically relevant 3D and adherent cultures is essential for elucidating dynamic cellular processes in stem cell biology and organoid development. Conventional imaging techniques often require invasive labeling or are limited in capturing transient phenotypes with minimal environmental disturbance. Here, we present a novel imaging platform optimized for continuous, non-invasive time-lapse acquisition of complex, self-organizing cellular systems. Our technology supports high-resolution monitoring of cellular dynamics in situ, enabling quantitative analysis of morphological and motility features without perturbing the native culture environment.

We demonstrate the utility of this platform in two experimental contexts: (1) organoid cultures, where we quantitatively tracked single-organoid growth dynamics, morphological remodeling, and motility under varied stress conditions; and (2) adherent stem cell populations, enabling single-cell tracking of motility patterns, cluster formation, and early differentiation-associated phenotypes. Preliminary data underscore the platform's capacity to extract biologically relevant metrics that complement conventional assays, facilitating detailed phenotypic profiling over extended culture periods.

These results validate the platform's applicability for high-content phenotyping and underscore its potential integration into workflows for organoid-based disease modeling, drug screening, and stem cell research. Ongoing development focuses on refining computational analysis modules tailored to stem cell and organoid applications, aiming to empower the research community with robust tools for dynamic cellular system interrogation.

Abstract No. P070

Investigating the role of LZTR1 in noonan syndrome-associated brain development using iPSC-derived models

Ana Trpchevska[1,*], *Belén Carriquí Madroñal*[1], *Anastasia Geladaris*[1,2], *André Fischer*[3,4], and *Lukas Cyganek*[1,5]

[1] Fraunhofer Institute for Translational Medicine and Pharmacology, Translational Neuroinflammation and Automated Microscopy, Göttingen
[2] Inst. Neuropathology, Uni. Medical Center, Georg August Uni., Göttingen
[3] Dep. Psychiatry & Psychotherapy, Uni. Medical Center (UMG), Georg-August-Uni., Göttingen
[4] German Center for Neurodegenerative Diseases (DZNE), Göttingen
[5] Stem Cell Unit, Dep. Cardiology & Pneumology, Uni. Medical Center, Göttingen

*Presenting author

Noonan Syndrome (NS) is a multisystemic, genetic disorder mostly characterized by severe congenital heart defects; however, many patients also present with a varying severity of developmental delays and intellectual disabilities (Wingbermühle et al. 2022). The molecular mechanisms underlying these cognitive manifestations are still not well understood, particularly in patients with biallelic variants in the gene *LZTR1*, which result in an absent or non-functional protein. LZTR1 is a well-established negative regulator of the RAS-MAPK pathway (Steklov et al. 2018), a signaling pathway crucial in regulating key developmental processes, therefore, its loss leads to hyperactivation of RAS-MAPK signaling with further potential consequences for neuronal development.

Using iPSC-derived cortical excitatory and inhibitory neurons and 3D brain organoid models, we investigated the effect of LZTR1 deficiency on RAS-MAPK pathway activity and neuronal development, maturation, and signaling.

In 2D neuronal cultures, we observed an accumulation of RAS GTPases MRAS and RIT1, as well as an increased pERK/ERK ratio, both consistent with increased RAS-MAPK signaling. At the 3D level, our data revealed an initial increase in mature MAP2-positive neurons in early day 30 *LZTR1* knockout (KO) organoids, suggesting premature neuronal maturation. However, by day 60, KO organoids displayed a slightly reduced population of mature neurons relative to the control organoids, indicating a possible failure to sustain long-term maturation. Furthermore, morphological analysis at day 50 demonstrated an expanded ventricular zone in KO organoids, pointing to altered neurodevelopment.

While preliminary, the results emphasize a possible way *LZTR1* mutations might contribute to the neurodevelopmental phenotypes seen in NS patients. Further studies with additional organoid batches, together with functional assays of neuronal and network activity, will be essential to more accurately define the role of LZTR1 in neuronal development. Ultimately, this work could impact development of therapeutic strategies aimed at modulating RAS-MAPK pathway activity to improve cognitive outcomes in NS.

Wingbermühle, E. et al. (2022). Cognitive Phenotype and Psychopathology in Noonan Syndrome Spectrum Disorders through Various Ras/MAPK Pathway Associated Gene Variants. Journal of clinical medicine, 11(16), 4735. https://doi.org/10.3390/jcm11164735

Steklov, M. et al. (2018). Mutations in LZTR1 drive human disease by dysregulating RAS ubiquitination. Science (New York, N.Y.), 362(6419), 1177–1182. https://doi.org/10.1126/science.aap7607

Abstract No. P071

Immune checkpoint molecules, the secrets of stem cell privilege in the lung

Roxana Maria Wasnick[1,], Nicolai Schmidt[2], Huijuan Wang[3], Deepesh Dhakad[1], Janine Gote-Schniering[3,4], Mareike Lehmann[5], Herbert Schiller[1], Ali Önder Yildirim[1,6]*, and *Andreas Günther[2]*

[1] Helmholtz Center Munich, Member of the German Center for Lung Research (DZL)
[2] University of Giessen and Marburg Lung Center (UGMLC), Member of the German Center for Lung Research (DZL)
[3] University of Bern, Switzerland
[4] Bern University Hospital, Switzerland
[5] Philipps-University Marburg, UGMLC
[6] Institute of Lung Health and Immunity, Helmholtz Munich

*Presenting author

The lung is an environmental barrier organ that is monitored extensively by the immune system. In Idiopathic Pulmonary Fibrosis (IPF), a progressive, fatal disease characterized by chronic epithelial injury and defective repair, epithelial cells in aberrant epithelial states accumulate. In contrast, the stem cell compartment is exhausted. We previously demonstrated in mouse and human the existence of a quiescent epithelial stem cell population expressing high levels of PD-L1(1). Here, we propose that the expression of the innate immune checkpoint molecule CD47 plays an essential role in the immune-monitoring and protection of epithelial stem cells and transitional states, as vulnerable non-homeostatic states. We hypothesize that CD47 is preferentially expressed by epithelial stem cells in the lung, thus conferring an immune-privileged status. Flow cytometry analyses of lung epithelial cells from IPF patients and bleomycin-treated mice demonstrate a general decline of cells surface CD47 in alveolar and non-alveolar epithelial cells. Simultaneously, in the mouse lung epithelium there is an expansion of a lineage diverse, $CD47^{high}$ population of self-renewing epithelial cells exhibiting a high clonogenic frequency and multi-lineage differentiation potential. Organoid assays demonstrate this population has a tenfold increase in clonogenic frequency, and it gives rise to all epithelial lineages in the lung epithelium. At the single-cell level, CD47mRNA expression is limited to a subpopulation of alveolar, bronchiolar and transitional K8/ADI epithelial cells, indicating active immune protection maintenance. The bulk epithelial population is endowed with a limited pool of CD47 protein distributed between daughter cells with each proliferation cycle. We thus propose that CD47 functions as a rheostat integrating the proliferative history and self-status of epithelial cells in the lung.

Ahmadvand N, Carraro G, Jones MR, et al. Cell-Surface Programmed Death Ligand-1 Expression Identifies a Sub-Population of Distal Epithelial Cells Enriched in Idiopathic Pulmonary Fibrosis. Cells. 2022;11(10):1593. doi:10.3390/cells11101593

Ahmadvand N, Khosravi F, Lingampally A, et al. Identification of a novel subset of alveolar type 2 cells enriched in PD-L1 and expanded following pneumonectomy. Eur Respir J. 2021;58(5):2004168. Published 2021 Nov 4. doi:10.1183/13993003.04168-2020

Abstract No. P072

iPSC-derived choroid plexus organoids for modeling Huntington's disease

Wenqing Xu[*], *Stephanie Le*, *Selene Lickfett*, *Felix Distelmaier*, and *Alessandro Prigione*

Heinrich Heine University, Düsseldorf

*Presenting author

Introduction: Huntington's Disease (HD) is a lethal neurodegenerative disease caused by abnormal CAG repeat expansion in *HTT* gene. Although the most common age of onset is in mid-life, recent publications also demonstrated brain developmental impairment. Choroid plexus (ChP) plays an important role in neural development by producing cerebral spinal fluid (CSF). Also, ChP forms blood–cerebrospinal fluid barrier (BCSFB) that regulates substance exchange. In this study, we plan to use induced pluripotent stem cell (iPSC)-derived ChP organoids to identify changes in ChP, CSF and BCSFB in HD during brain development stage.

Material/Methods: We established ChP organoid differentiation protocol based on Lancaster et al[1] in 2020. The mature ChP organoids were characterized by ChP and CSF markers expression. BCSFB integrity was evaluated based on cell junction markers and FITC-dextran permeability assay. Edited iPSCs with 70Q/70Q in *HTT* were previously generated using CRISPR/Cas9 to model HD. Control iPSCs with 21Q/21Q in *HTT* and HD iPSCs are planned to be differentiated into ChP organoids and the aforementioned phenotypes would be tested.

Results: ChP organoids were successfully generated with visible cysts filled with CSF-like fluid (iCSF). ChP markers TTR and CLIC6 were confirmed in the ChP organoids. Using WB, we found Clusterin and TTR expression in the iCSF, culture medium as well as the whole ChP organoids. Regarding BCSFB, positive staining of Claudin-1 and ZO-1 were identified in ChP epithelium, and FITC-dextran permeability assay confirmed limited dextran crossing the ChP epithelium. The *HTT* genotype of control and HD iPSCs line were confirmed by gel electrophoresis and Sanger sequencing. Control and HD ChP organoids are currently in culture and waiting to be characterized.

Discussion: Using iPSC-derived ChP organoids, we aim to excavate neural developmental changes in HD including CSF production and BCSF integrity, which would give new insights into the pathogenesis of HD in early stages.

Pellegrini L et al, Science, 2020.

Poster session II: P073 – P144

P073 – P081: Computational stem cell biology & systems biology

P073 Transcriptomic signatures of clonally related cell pairs uncover molecular control of fate stabilization
Ida Büschel

P074 Disentangling transcription factor programs for direct reprogramming with generative modeling
Leon Hafner

P075 A study of the genetic circuits of intestinal stem cell differentiation using in vivo CRISPR perturbation and single-cell RNA-seq
Anastasiia Horlova

P076 Dynamical modeling-informed interpretation of Ki67 positivity
Adrien Jolly

P077 Generative machine learning for cell programming
Dominik Klein

P078 The digital bone marrow niche – a spatio-temporal in silico model of hematopoietic (stem) cells in their bone marrow microenvironment
Osman Abhimata Nugraha

P079 A computational pipeline to characterize non-human primate iPSCs according to ISSCR standards
Felix Pförtner

P080 Snapshot-based identification of mutant fitness and lineage bias in clonal hematopoiesis of indeterminate potential via a three-compartment model
Daniele Scarcella

P081 Topological evaluation and comparative application of gene regulatory network inference from single-cell sequencing data
Marco Stock

Abstract No. P073

Transcriptomic signatures of clonally related cell pairs uncover molecular control of fate stabilization

Ida Büschel[1,2,*], *Keerthana Arun*[3], *Yogesh Goyal*[3], and *Carsten Marr*[1,4]

[1] Helmholtz Munich, Neuherberg
[2] Biomedical Center, Faculty of Medicine, LMU Munich, Planegg-Martinsried
[3] Feinberg School of Medicine, Northwestern University, Chicago, US
[4] LMU University hospital, Munich

*Presenting author

Diverse cell fates emerge from a balance between variability and stability: While asymmetric cell divisions are thought to generate fate diversity, clonally related cells often remain transcriptionally similar for extended periods, a phenomenon sometimes described as “sister-cell memory.”

To investigate this interplay, we analyzed clonally related pairs from three published lineage and time resolved single-cell RNA sequencing datasets. Cells were captured at two time points during stem cell differentiation, at early fate commitment and after progression into distinct lineages. Across both stages, clonally related cells were consistently more transcriptionally similar than unrelated pairs, reflecting inheritance of the parental cell state. Importantly, similarity decreased with increasing clone size, as stochastic differences accumulate and potentially coordinated asymmetric divisions generate fate diversity.

By directly comparing clonally related pairs, we identified transcriptional signatures that distinguish clone-neutral genes, where the variability between clonally related cells does not significantly differ from that between randomly paired cells, from clone-specific genes, which retain greater similarity in clonally related cell pairs. Comparison with “memory genes”, as identified in independent heritability experiments, suggests that these clone-specific genes may reflect mechanisms that help reinforce emerging cell identities.

Unlike conventional approaches that rely on differentially expressed or highly variable genes between predefined cell types, our framework does not require prior annotations and can reveal informative differences even from a single time point.

Analyzing clonally related cell pairs is a powerful tool for probing the duality of cell fate decisions. Asymmetry may introduce diversity, but memory in clonally related cells points toward regulatory features that potentially stabilize fates during lineage commitment.

Goyal et al., “Diverse Clonal Fates Emerge upon Drug Treatment of Homogeneous Cancer Cells.”

Filipczyk, Marr, Hastreiter et al., "Network plasticity of pluripotency transcription factors in embryonic stem cells."

Abstract No. P074

Disentangling transcription factor programs for direct reprogramming with generative modeling

*Leon Hafner**, *Mariia Minaeva*, *Dominik Klein*, and *Fabian J. Theis*

Institute of Computational Biology, HMGU, Hellmholtz Munich

*Presenting author

Transcription factor-induced reprogramming enables the direct transition between differentiated cell types without passing through a pluripotent state. This avoids risks associated with stem cell-based approaches, such as tumorigenicity and uncontrolled differentiation, while offering a comparatively rapid way to generate immune cells suitable for therapeutic use. However, identifying efficient transcription factor combinations remains a major challenge due to the intractable space of possible factors and their concentrations. Only very few cocktails have been identified so far, often with limited efficiency, and their discovery is highly resource-intensive, leaving many immune lineages directly inaccessible (Rosa *et al.* 2018).

In this work, an adapted version of CellFlow (Klein *et al.* 2025), a flow-matching-based generative modeling framework for single cells, is trained on a single-cell RNA-seq dataset of human embryonic fibroblasts subjected to highly multiplexed transcription factor perturbations. Our model disentangles the underlying cellular transitions of the reprogramming landscape by learning how transcription factor treatments shape gene expression, and generates predicted gene expression profiles for arbitrary TF combinations.

We were able to validate the performance of the model on previously established and experimentally confirmed TF combinations that reprogram fibroblasts into dendritic cells (Rosa *et al.* 2018). Building on this, we optimized previously established transcription factor combinations and identified new ones for reprogramming fibroblasts into various dendritic cell types. By analyzing the gene expression profiles of the generated cells, we demonstrate that these correspond to medically relevant phenotypes and cell states.

Overall, this framework establishes a systematic and computationally feasible strategy to uncover transcription factor programs for direct lineage reprogramming. If experimentally validated, these predictions would provide access to previously unattainable immune states and have the potential to accelerate the development of next-generation immunotherapies.

Rosa FF, Pires CF, Kurochkin I, Ferreira AG, Gomes AM, Palma LG, et al. Direct reprogramming of fibroblasts into antigen-presenting dendritic cells. Sci Immunol. 2018;3. doi:10.1126/sciimmunol.aau4292

Klein D, Fleck JS, Bobrovskiy D, Zimmermann L, Becker S, Palma A, et al. CellFlow enables generative single-cell phenotype modeling with flow matching. bioRxiv. 2025. doi:10.1101/2025.04.11.648220

Abstract No. P075

A study of the genetic circuits of intestinal stem cell differentiation using in vivo CRISPR perturbation and single-cell RNA-seq

Anastasiia Horlova[1,*], *Siamak Redhai*[2], *Stefan Peidli*[1], *Michael Boutros*[2,3], and *Wolfgang Huber*[1]

[1] EMBL Heidelberg
[2] The German Cancer Research Center (DKFZ), Heidelberg
[3] Heidelberg University

*Presenting author

Understanding how genetic networks enable animals to develop and maintain complex tissues is a central goal of biological research. The Drosophila midgut is a well-established and important model system that has enabled many discoveries that generalize to other animals, including humans. The gut has remarkable plasticity, high self-renewal, and is prominently exposed to environmental factors.

Our research investigates how intestinal stem cells (ISCs) contribute to the generation of differentiated cell types in the Drosophila midgut. We use single-cell transcriptomic data from in vivo CRISPR gene knockdown experiments that specifically affect Drosophila melanogaster ISCs, in their native context within healthy adult animals. Our goal is to systematically examine the consequences of single gene perturbations on cellular behaviour. The dataset comprises sc-RNAseq transcriptomic profiles of 427,549 cells from 61 replicate experiments of 28 unique genetic perturbations.

In our analysis workflow we preprocess and filter the data to eliminate low-quality cells. We pool the wild type samples and integrate them via the Harmony algorithm. We use clustering and marker gene expression to annotate cell-types. We annotate perturbed cells using a reference-based k-nearest neighbors classifier. We examine differential gene expression via pseudo-bulk DESeq2 analysis, and evaluate changes in cell-type proportions via compositional analysis. We use lineage inference to identify transcription factors involved in differentiation. Gene set enrichment analysis will help identify pathways associated with ISC differentiation. Shared pathways, transcriptional factors, and gene expression changes across perturbations will be analyzed to reveal underlying genetic network dynamics. We use dimensionality reduction and visual summaries (e.g., UMAPs, heatmaps, volcano plots) to support biological interpretation of the data.

Ultimately, this work aims to offer novel insights into genetic network dynamics, advancing our understanding of cellular differentiation and organ development.

Abstract No. P076

Dynamical modeling-informed interpretation of Ki67 positivity

Adrien Jolly[1,*], *Michael Rieger*[1], and *Florian Büttner*[1, 2]

[1] Goethe University Frankfurt, University Hospital, Department of Medicine 2 - Hematology and Oncology, Frankfurt am Main
[2] DKTK - German Cancer Consortium, Frankfurt

*Presenting author

Ki67 is a protein with multiple functions during the cell cycle, but it is mostly known as a proliferation marker. Both in cell imaging and flow cytometry, it is usually used as a binary proliferation versus quiescence marker, where a fixed level of fluorescence intensity separates cycling and quiescent cells.

It is now established that the protein is degraded continuously in both G1 and G0 phases of the cell cycle and produced in S and G2M [1]. However, the consequences of this expression pattern for the interpretation of binary (Ki67 positive or negative fractions) remain largely unaccounted for, leading to systematic errors, with potentially massive over or underestimation of the quiescent fraction depending on the length of the G1 phase and the degradation kinetics of Ki67.

In this work, we describe quantitatively how the percentage of Ki67 positive cells at steady state varies depending on the cell cycle dynamics. For this purpose, we use a realistic population-level mathematical model of the cell cycle that we combine with a literature-informed model of Ki67 kinetics. Briefly, we assume that Ki67 negative cells become positive upon entering the S phase, and that Ki67 fluorescence decays continuously in G0/G1 from a uniform starting level after mitosis. Consequently, in our model, Ki67 positive cells in G0/G1 rapidly turn negative following a fixed delay after division.

This model allows us to explore various scenarios and determine how Ki67 positivity is expected to change upon perturbations of the cell cycle. We find notably that an increase in Ki67 positivity in G0/G1 for a given cellular system is generally indicative of a higher global proliferation, but not necessarily of the activation of quiescent cells. Our work provides guidelines for both experimentalists and clinicians to interpret changes in Ki67 positivity.

[1] Miller I, Min M, Yang C, Tian C, Gookin S, Carter D, et al. Ki67 is a graded rather than a binary marker of proliferation versus quiescence. Cell Rep. 2018;24(5):1105–1112.e5. https://doi.org/10.1016/j.celrep.2018.06.110

Abstract No. P077

Generative machine learning for cell programming

Dominik Klein[1,*], *Jonas Fleck*[2], *Gray Camp*[2], and *Fabian Theis*[1]

[1] Institute of Computational Biology, Helmholtz Munich
[2] Institute of Human Biology, Basel, Switzerland

*Presenting author

Programming cellular states remains a major challenge, given the vast combinatorial space of modulators, dosages, and exposure times. Exhaustively exploring this protocol space experimentally is infeasible. As such, in silico prediction of cell fates offers a promising strategy to prioritize experimental conditions and accelerate cell fate engineering. However, this requires expressive machine learning models capable of capturing the inherent heterogeneity of cellular states within complex tissues. We present CellFlow, a generative model based on flow matching that predicts cellular responses to perturbations. CellFlow enables accurate phenotype prediction in neuronal reprogramming and extends beyond traditional low-dimensional assays to perform in silico screens in 2D neuron models and 3D brain organoid models. Altogether, CellFlow establishes a new paradigm for optimizing cell fate engineering using powerful generative modeling techniques.

Abstract No. P078

The digital bone marrow niche – a spatio-temporal in silico model of hematopoietic (stem) cells in their bone marrow microenvironment

*Osman Abhimata Nugraha**, *Thomas Zerjatke*, *Ingmar Glauche*, and *Ingo Roeder*

Technische Universität Dresden

*Presenting author

Hematopoietic stem cell (HSC) maintenance in the bone marrow, including aspects like self-renewal, differentiation, proliferation, and quiescence, critically depends on the local microenvironment, known as the stem cell niche. Dysregulation of this niche is known to contribute to hematological malignancies. However, the spatial complexity of the bone marrow hampers a full understanding of niche-driven HSC regulation and its relation to disease progression.

To address this, we are developing a 3D spatio-temporal *in silico* model based on the Cellular Potts Model (CPM) implemented in *Morpheus*. In this framework, HSCs and progenitor cells are represented as individual agents whose movements and behaviors evolve in space and time through a combination of deterministic and stochastic rules. These agents interact with each other, with niche components, and with signaling molecules within a particular 3D domain that represents the geometry of the niche (e.g., vasculature). This so-called "scaffold" can be derived from microscopy data or artificially generated via simulation of reaction-diffusion equations. Simulation outputs are analyzed to produce statistical measures that can be directly compared with experimental data.

At the current stage, our model incorporates two reversible HSC states, namely "active" (proliferating) and "inactive" (quiescent), thereby extending a previous model into a spatial context. Preliminary simulations in artificially generated scaffolds demonstrate distinct HSC population dynamics and residence time distributions depending on scaffold structure. These findings suggest that structural changes in the niche can alter HSC behavior in the bone marrow and may provide a starting point to better understand aging- or disease-related alterations in the microenvironment.

We aim to implement an iterative modeling approach in which simulation outputs are continuously compared with experimental observations, and the model is refined accordingly. Through this effort, we hope to advance mechanistic insights into spatial aspects of niche regulation in hematopoiesis and its dysregulation in aging and disease.

Starruß, J., de Back, W., Brusch, L. & Deutsch, A. Morpheus: a user-friendly modeling environment for multiscale and multicellular systems biology. Bioinformatics 30, 1331–1332 (2014).

Roeder, I. & Loeffler, M. A novel dynamic model of hematopoietic stem cell organization based on the concept of within-tissue plasticity. Exp Hematol 30, 853–861 (2002).

Abstract No. P079

A computational pipeline to characterize non-human primate iPSCs according to ISSCR standards

*Felix Pförtner**, *Antonia Keßler*, *Wolfgang Enard*, and *Ines Hellmann*

LMU, Munich

*Presenting author

The ISSCR standards for human stem cell lines, have significantly advanced the field. Many of the standard approaches for the 'Basic Characterization' of human cell lines can be directly applied to non-human primate (NHP) iPSC lines without modification. However, for the ISCCR standards for cell line authentication and the characterization of 'Pluripotency and the Undifferentiated State', existing protocols for human cell lines are unlikely to work for NHPs, as many of the resources, have been specifically developed and optimized for human iPSCs.

To address these challenges, we are developing resources and methods specifically tailored to the ISSCR-compliant characterization of NHP iPSC lines, with a focus on cynomolgus. Our approach leverages our RNA-seq library preparation method, prime-seq 2, along with its accompanying computational analysis pipeline, zUMIs, to authenticate cell lines, characterize their undifferentiated state and assess their pluripotency. Prime-seq 2 data can be utilized to achieve these goals, providing a robust measure of pluripotency and establishing a reliable framework for characterizing iPSC lines from other NHP species. Building on zUMIs, we are wrapping the workflow into a user-friendly computational pipeline.

Pförtner, Felix, Eva Briem, Wolfgang Enard, and Daniel Richter. 2025. "Improving RNA-Seq Protocols." bioRxiv. https://doi.org/10.1101/2025.08.20.671269.

Parekh, S., C. Ziegenhain, B. Vieth, W. Enard, and I. Hellmann. 2018. "zUMIs - A Fast and Flexible Pipeline to Process RNA Sequencing Data with UMIs." Gigascience 7 (6). https://doi.org/10.1093/gigascience/giy059.

Abstract No. P080

Snapshot-based identification of mutant fitness and lineage bias in clonal hematopoiesis of indeterminate potential via a three-compartment model

Daniele Scarcella[1,*], *Ivan Kukuljan*[1], *Linus J. Schumacher*[2], and *Carsten Marr*[1,3]

[1] Helmholtz Munich, Neuherberg
[2] University of Edinburgh, UK
[3] LMU University hospital, LMU Munich

*Presenting author

Clonal hematopoiesis of indeterminate potential (CHIP) is frequent in ageing and associated with mutations in DNMT3A, TET2, ASXL1, JAK2, among others, with links to myeloid malignancy and cardiovascular disease. Yet we still lack a clear picture of where, along the HSPC hierarchy, mutant advantage is realised and how lineage bias emerges across compartments.

We introduce a compact compartment model that follows a mutant clone through three key pools: HSCs (H), myeloid-committed (M) and lymphoid-committed (L). The model distils mutant behaviour into two interpretable levers: a fitness term that accelerates expansion within HSCs, and a lineage-bias term that redistributes output between myeloid and lymphoid branches.

First, we show in simulations that single-time snapshots can identify both fitness and lineage bias. With HSCs alone, a single-time snapshot can only identify the product of fitness and clone age, but adding the myeloid- and lymphoid-committed compartments breaks this non-identifiability. Second, we provide a practical inference workflow: MCMC to estimate fitness, lineage bias, and (optionally) clone age from one snapshot, plus a sensitivity analysis that perturbs myeloid and lymphoid turnover rates to deliver robust intervals for lineage bias. Applied to published single-cell data (Jakobsen et al., 2024), the model recovered patient-specific fitness and suggested lineage bias in one DNMT3A and one TET2 case.

Together, these results clarify where mutant advantage sits in the hierarchy, quantify to what extent fitness and lineage bias contribute to clonal expansion, and furnish a snapshot-based tool for early risk stratification and hypothesis testing in CHIP.

Jakobsen, N. A. et al. Selective advantage of mutant stem cells in human clonal hematopoiesis is associated with attenuated response to inflammation and aging. Cell Stem Cell, 31(S1934-5909(24)00207), doi: https://doi.org/10.1016/j.stem.2024.06.019

Abstract No. P081

Topological evaluation and comparative application of gene regulatory network inference from single-cell sequencing data

Marco Stock[*], *Matteo Zambon*, and *Antonio Scialdone*

IES - Institute of Epigenetics and Stem Cells, Helmholtz Munich

*Presenting author

Optimizing Gene Regulatory Network (GRN) inference from single-cell RNA sequencing (scRNA-seq) data presents a remaining challenge in computational biology. Despite the availability of extensive scRNA-seq datasets, existing machine learning-based methodologies have demonstrated limited eIicacy in accurately modelling GRN complexities, particularly when assessed through binary classification of graph edges. Our study introduces a novel perspective by evaluating the capacity of these methods to accurately reconstruct GRN graph topologies, revealing inherent topological biases in current predictive algorithms.

We finally present our application of GRN inference to benchmark newly developed in vitro models of human embryonic development against corresponding in vivo systems. This comparative analysis aims to advance our understanding of in vitro models of embryonic development through computational modelling.

Stock et al., Topological benchmarking of algorithms to infer gene regulatory networks from single-cell RNA-seq data, Bioinformatics, Volume 40, Issue 5, May 2024, btae267, https://doi.org/10.1093/bioinformatics/btae267

Stock et al., Leveraging prior knowledge to infer gene regulatory networks from single-cell RNA-sequencing data, Mol Syst Biol (2025) 21: 214 - 230, https://doi.org/10.1038/s44320-025-00088-3

P082 – P124: Stem cells in disease modeling & drug development

P082 Organ-specific expression data steers choice of laminin for in vitro studies in biologically relevant drug discovery and development platforms
Chiara Annunziata

P083 Exploring schizophrenia pathogenesis: hiPSC models and genetic insights
Ágota Apáti

P084 Functional and molecular profiling of iPSC-derived cell types from bipolar disorder patients in Spanish multiplex families
Ankita Kumar Bhamidipati

P085 Allelic switching underlies compensation in ACTB-deficient iPSCs
Stephanie Binder

P086 Modeling the impact of plastic pollution on human neurodevelopment and neuroinflammation
Nelli Blank-Stein

P087 Utilising urine-derived stem cells for disease modelling and drug screening
Kazette Yuen Yu Chan

P088 A human iPSC model of tauopathies engineered for 4R Tau isoform expression endogenously develops late-stage neuronal Tau pathology
Angelika Dannert

P089 Impact of CNTNAP2 deficiency on dendritic arborization of human neurons
Saloia Es Sbai

P090 Inflammation-mediated modulation of synaptic function in SHANK3-deficient human neuronal networks
Christian Felski

P091 An innovative human iPSC cardiomyocyte platform to accelerate cardiac drug discovery and target validation
Benjamin Fischer

P092 Adult ADHD, PARK2 copy number variations and the blood-brain barrier: Metabolic studies in hiPSC-derived in vitro models
Markus Glaser

P093 Minimizing observational bias in cell research: Applying low-impact imaging technologies for reliable datasets
Thomas Gohl

P094 Automated high-throughput wound-healing assay for quantitative analysis of cell migration
Aleks Guledani

P095 Developing antisense oligonucleotide therapies for SCA3 using iPSC-derived neural models
Stefan Hauser

P096 iPSC-derived cortical brain organoids and neurons for the study of pathomechanisms and gene therapy approaches in Leigh syndrome
Sonja Heiduschka

P097 Development of iPSC-derived models for succinic semialdehyde dehydrogenase deficiency and their cellular and molecular profiling
Samuel Hofmann

P098 Rewiring the pre-cancer landscape: KRAS-driven secretome involving TNFα shapes the pancreatic niche
Meike Hohwieler

P099 Development of autologous iPSC-based immune systems-on-chip for infectious disease research
Reyk Horland

P100 A fully human glioblastoma-in-brain-spheroid model enables high-throughput compound screening
Sandra Horschitz

P101 Leigh syndrome neurodevelopmental dysfunction modeled in two and three dimensions
Caleb Jerred

P102 Development of a high-throughput screening method for drug target discovery in schizophrenia by means of utilizing co-cultured hiPSC-derived cortical neurons and microglia
Jessica Jung

P103 GTP cyclohydrolase I deficiency leads to mitochondrial impairment in iPSC-derived neural progenitor cells and Ngn2-induced neurons
Sabine Jung-Klawitter

P104 Beyond plastic: Engineering soft environments with ultra-high viscosity alginate for hiPSC-derived neurons in vitro
Isabell Karnatz

P105 Modelling PURA syndrome in cerebral organoids uncovers cell type misdistribution as likely cause for symptoms in patients
Carolin Ketteler

P106 Advancing and modulating human heart-forming organoids (HFOs) for drug testing
Felix Kleemiß

P107 ALS causative KIF5A mutations lead to translational impairment and nucleolar stress in hiPSCs derived motor neurons
Philipp Koch

P108 Identification and mitigation of major confounders in iPSC-based psychiatric disease modeling
Tamara Krutenko

P109 Kidney organoids as a human iPSC-derived in vitro model for Fabry nephropathy
Sandra D. Laufer

P110 Advanced functional assays reveal co-culture and substrate-driven maturation and patient-specific differences in hiPSC cardiomyocytes and neurons
Marta Lemme

P111 A novel KIT D816V mutated hiPSC-mast cell model with IgE-dependent activation
Yanyan Luo

P112 Modeling PURA syndrome: impact of PURA knockout and F233del mutation on neural progenitor cells
Silvia Marcato

P113 Human 3D neuromuscular organoids to model TDP-43 related mechanisms in ALS
Aylin Nebol

P114 Repeated rhinovirus infections drive epigenetic memory and senescence
Wasifa Nurieva

P115 Patient iPSC-derived microglia reveal disease-associated changes in BPAN
Gamze Özata

P116 Development of a hiPSC-based platform for high-throughput drug screening and disease modeling in hereditary pulmonary arterial hypertension
Clara Luise Plötner

P117 Single-cell profiling of striatal organoids derived from Leigh syndrome patients indicates an early imbalance of excitatory and inhibitory neurons during neurodevelopment
Martina Podlesnic

P118 High throughput screening for compounds allowing readthrough of premature termination codons based on genetically engineered Primary Ciliary Dyskinesia patient specific hiPSCs
David Puertollano Martín

P119 Genetic screen to identify lipid metabolism regulators using hiPSC-derived hepatocytes
Merrit Romeike

P120 Psilocin fosters neuroplasticity in iPSC-derived human cortical neurons
Malin Schmidt

P121 Network hyperexcitability and dopaminergic neuron loss in a novel A30P-driven Parkinson's midbrain organoid model
Kea Aline Schmoll

P122 Mitochondrial DNA editing enables the development of iPSC models of maternally inherited Leigh syndrome
Isabella Tolle

P123 Human iPSC-derived BBB model for CNS-targeted PROTAC discovery
Arjen Weller

P124 Modeling and reversing Alzheimer's-associated microglial states using iPSC-derived cells and disease-linked gene signatures
Bastian Zimmer

Abstract No. P082

Organ-specific expression data steers choice of laminin for in vitro studies in biologically relevant drug discovery and development platforms

*Chiara Annunziata**, *Sam Hobson*, *Elias Uhlin*, *Sandra Falci*, *Noah Fereydouni*, *Evan Lee Graham*, *Malin Kele*, and *Therese Kallur*

BioLamina, Sundbyberg, Sweden

*Presenting author

The Drug Discovery and Development (DDD) process employs various cell culture platforms to evaluate the safety and efficacy of lead compounds. With advancements in stem cell research, there is a growing shift from traditional 2D cultures to more complex, patient-derived models. Human pluripotent stem cells (hPSCs) require an extracellular matrix (ECM) for maintenance, proliferation, and differentiation. Laminins, a major ECM protein family found in basement membranes, are critical to stem cell niches. Despite their increasing use in hPSC studies, standardized protocols incorporating biologically relevant ECMs in DDD platforms remain underdeveloped.

We analyzed protein and gene expression patterns of laminin isoforms in four key tissues often used in DDD - heart, liver, kidney, and central nervous system—revealing tissue- and cell-type specificity. The kidney exhibited unique isoform expression compared to other tissues. Translating this data to in vitro systems, we cultured specific cell types on tissue-relevant laminin substrates. Results showed significantly enhanced proliferation and cell functionality. For example, laminin-coated cultures produced 43-times more dopaminergic neurons (1) and over 80% functional cardiovascular progenitor cells, compared to 50% with standard substrates (2). Furthermore, when Biolaminins is integrated into microfluidic systems, it enables the recreation of dynamic in vivo-like environments. These platforms enhance differentiation, reproducibility, and maturation of hPSC-derived cells, addressing current limitations in DDD.

Our findings highlight the importance of using tissue-specific laminin isoforms to closely mimic native ECM environments in vitro. By aligning in vivo expression data with in vitro applications, it is possible to develop standardized and functional cell-based models. This approach supports the transition from animal models to more predictive human-based platforms, such as organ-on-a-chip systems, ultimately improving the physiological relevance of preclinical testing and accelerating drug development.

Kirkeby et al. Predictive Markers Guide Differentiation to Improve Graft Outcome in Clinical Translation of hESC-Based Therapy for Parkinson's Disease. Cell Stem Cell. 2017

Yap et al. Generation of Post-infarct Human Cardiac Muscle by Laminin-Promoted Cardiovascular Progenitors. Cell Rep. 2019

Abstract No. P083

Exploring schizophrenia pathogenesis: hiPSC models and genetic insights

Csongor Tordai[1], *Edit Hathy*[2], *Katalin Vincze*[1], *Kiara Farkas*[1], *Ádám Póti*[1], *Dávid Szüts*[1], *János M. Réthelyi*[3], and *Ágota Apáti*[1,*]

[1] Institute of Molecular Life Sciences, HUN-REN RCNS, Budapest, Hungary
[2] Molecular Psychiatry Research Group, Semmelweis University, Budapest, Hungary
[3] Department of Psychiatry and Psychotherapy, Semmelweis University, Budapest, Hungary

*Presenting author

Our work explores the viability of using human induced pluripotent stem cell (hiPSC)-based in vitro models to study schizophrenia (SCZ), focusing on two patient cases with de novo mutations. We established and validated patient-specific hiPSC lines and utilized CRISPR genome editing along with parental cell reprogramming to create robust genetic controls. By implementing a hippocampal differentiation protocol, we successfully differentiated hiPSCs into neural progenitor cells (NPCs) and dentate gyrus granule cells (DGGCs), which developed into functional neurons, as confirmed through calcium imaging and electrophysiological assessments. Our results revealed significant transcriptomic differences between SCZ and control cell lines, particularly in genes associated with neuronal differentiation and synaptic function, supporting the neurodevelopmental hypothesis of SCZ. Both patient cases exhibited altered glutamatergic signaling, consistent with established SCZ characteristics. These findings provide evidence of cell-autonomous phenotypes in SCZ and highlight the influence of rare genetic variants on the disorder, underscoring the importance of integrating transcriptomic and functional data to investigate disease mechanisms. Future research should involve larger patient cohorts to determine whether distinct genetic mutations converge on fundamental biological systems, particularly synaptic pathology and glutamatergic pathways, potentially revealing shared mechanisms underlying SCZ and informing targeted therapeutic strategies.

Funding of this study was provided by the Hungarian Brain Research Program (Nemzeti Agykutatási Program, NAP) grants NAP-B KTIA_NAP_13-2014-0011 and 2017-1.2.1-NKP-2017-00002 to JR and NAP-A KTIA_13_NAP-A-I/6 to ÁA , GINOP-2.1.1-15-2015-00369 to ÁA, and the Richter Gedeon Pharmaceuticals Téma Pályázat Grant No. RG-IPI-2016 TP10/015 to DS.

1: Hathy E et al. Stem Cell Res Ther. 2020 PMID: 33246498 2: Tordai C et al. Schizophr Res. 2024 PMID: 38290943. 3: Farkas KG et al. Int J Mol Sci. 2025 PMID: 40362679

Abstract No. P084

Functional and molecular profiling of iPSC-derived cell types from bipolar disorder patients in Spanish multiplex families

*Ankita Kumar Bhamidipati**, *Anne Hoffrichter*, *Tessa Fabian*, *Malin Schmidt*, and *Philipp Koch*

Central Institute of Mental Health, Medical Faculty Mannheim, Heidelberg University

*Presenting author

Background: Bipolar Disorder (BD) is a neuropsychiatric condition with a strong genetic basis. However, the mechanisms by which genetic risk factors influence gene expression, molecular pathways, and neural function remain poorly understood. This study utilises induced pluripotent stem cells (iPSC)-derived cell types to investigate disease-relevant cellular and molecular phenotypes.

Methods: We differentiated iPSC lines from four BD type 1 patients (from multiplex families) and four age- and sex-matched unrelated controls into cortical progenitors, neurons, and astrocytes. We performed transcriptomic profiling and high throughput immunocytochemistry (PerkinElmer/Revvity Opera Phenix) to assess differential gene expression and cellular phenotypic differences respectively between the BD and control groups.

Results: Within the cortical progenitors, our analysis revealed significant differential expression of neurodevelopment-associated genes, notably the collagen and ephrin gene families. In the BD iPSC-derived neurons, we found altered expression of protocadherin family genes and additional neuropsychiatric disorder-related genes including DISC1 and NRXN1. We also observed dysregulation of multiple potassium channel genes in both progenitors and neurons, which suggests a hyperexcitability phenotype. In the BD iPSC-derived astrocytes, we demonstrated elevated expression of ALDH1L1, S100ß, and AQP4, indicating increased reactivity or an altered maturation state. We are currently in the process of validating the above findings and correlating them with morphometric and electrophysiological alterations.

Conclusion: Our findings reveal transcriptomic and phenotypic alterations across different cell types in BD, implicating disrupted neurodevelopmental, excitability, and glial pathways. This integrative, cell-type-specific approach aims to uncover key pathways underlying BD pathophysiology, supporting mechanism-based therapeutic strategies.

Abstract No. P085

Allelic switching underlies compensation in ACTB-deficient iPSCs

Andrea Rossi, *Thomas Haarmann-Stemmann*, and *Stephanie Binder**

IUF - Leibniz research institute for environmental medicine, Düsseldorf

*Presenting author

Loss-of-function (LOF) mutations in *ACTB*, encoding β-Actin, are linked to developmental disorders with variable expressivity, suggesting the presence of compensatory mechanisms. However, the molecular basis of this compensation remains poorly understood.

To investigate this, we generated isogenic human iPSC models with heterozygous *ACTB* mutations using CRISPR/Cas9 and prime editing: either a premature termination codon (PTC) to retain the locus and generate mutant mRNA, or a full-locus deletion to eliminate both mRNA and protein. This design allowed us to distinguish whether compensation is triggered by mutant mRNA (via mRNA surveillance/feedback) or by the absence of protein. Complete knockouts were chromosomally unstable, but both heterozygous models were cytogenetically normal, enabling detailed analysis of compensatory responses.

In PTC mutants, we observed early upregulation of *ACTB* and gene paralogues, particularly *ACTG2*. In contrast, mutants with a full-locus deletion showed no immediate compensation, but gradually upregulated *ACTB* after multiple passages, suggesting a delayed, possibly selection-driven, adaptation. While consistent with genetic compensation, the exact mechanism underlying paralogue upregulation remains to be determined.

Surprisingly, total *ACTB* transcript levels in PTC mutants were comparable to wild-type iPSCs despite clear degradation of the mutant allele. Digital PCR and long-read sequencing revealed that this was due to robust upregulation of the remaining wild-type allele. Actinomycin-D time-courses showed no change in mRNA stability, while 4-thiouridine labeling and pre-mRNA analysis confirmed an increase in transcription from the wild-type allele.

This work highlights the value of engineered isogenic models for dissecting gene dosage effects and reveals allelic switching as a critical compensatory mechanism that may underlie phenotypic variability and resilience in a broad range of haploinsufficient genetic disorders.

Abstract No. P086

Modeling the impact of plastic pollution on human neurodevelopment and neuroinflammation

Nelli Blank-Stein[1,*], *Marina Mayer*[1], *Maria Francesca Viola*[1], *Omkar Suhas Vinchure*[2], *Jay Gopalakrishnan*[2], and *Elvira Mass*[1]

[1] Life & Medical Sciences Institute (LIMES), University of Bonn
[2] University Hospital, Friedrich Schiller University Jena

*Presenting author

Brain development is shaped by genetic programs and environmental exposures. Plastic pollution, driven by the widespread use of micro- and nanoplastics (MNPs), poses a growing environmental concern, yet its effects on neurodevelopment and neuroinflammation remain largely unknown. Given the regulatory role of microglia in brain development, we investigated how MNPs affect human brain development using iPSC-derived models, including brain organoid model systems with and without integrated microglia, as well as induced microglial-like cells (iMGLs). Single-cell RNA sequencing of MNP-exposed organoids revealed reduced neuronal and glial differentiation and increased stress-responsive neuronal populations, suggesting disrupted developmental trajectories. Proliferation assays confirmed disrupted cell cycle dynamics, and chronically exposed organoids exhibited significant size reduction. However, this growth deficit was partially rescued in organoids containing microglia, indicating a potential microglial role in buffering MNP-induced growth impairments. In iMGLs, live-cell imaging and flow cytometry demonstrated phagocytic uptake of MNPs without induction of a classical inflammatory response. Transcriptomic analysis showed upregulation of cellular stress and immune response pathways, alongside metabolic alterations. Metabolic profiling indicated a shift from glycolysis toward fatty acid biosynthesis, with increased expression of Acetyl-CoA Carboxylase 1 (ACC1) and decreased levels of key glycolytic and mitochondrial genes (GLUT1, ATP synthase, CPT1a, G6PD, SDHA, CytC, CD98), indicating altered energy homeostasis and metabolic reprogramming. Together, these findings suggest that plastic particle exposure disrupts neuronal differentiation and microglial function while reshaping metabolic programs. This iPSC-based platform offers a valuable tool to study environmental plastic exposure and provides insights into its impact on neurodevelopment and microglia–neuron interactions.

Abstract No. P087

Utilising urine-derived stem cells for disease modelling and drug screening

Kazette Yuen Yu Chan[1,*], *Karina da Costa Silveira*[2], *Alex Beke*[1], *Carry-lynn Soltys*[1], and *Peter Kannu*[1]

[1] University of Alberta, Canada
[2] University of Calgary, Canada

*Presenting author

Many rare human genetic disorders lack suitable models for studying the underlying mechanisms. Our lab focuses on skeletal dysplasias (SD), a group of childhood-onset disorders affecting bone and cartilage development. Diagnosing SD remains challenging because of variants of uncertain significance (VUS) in genetic test results. When a VUS is identified, the result neither confirms nor excludes the diagnosis. Further testing generally requires affected tissue, such as bone or cartilage, which is difficult to obtain.

Current research uses patient-derived stem cells commonly sourced from blood, skin, or bone marrow to create induced-pluripotent stem cells. As a non-invasive alternative, we propose using urine as a source. While existing studies demonstrate their regenerative ability, using urine-derived stem cells (UDSC) as a disease modelling and drug testing system remains underexplored. This project aims to leverage the differentiation potential of UDSC and explore the underlying biology of specific SD, functionally assess VUS, and test potential drug therapies.

Our preliminary results demonstrate the potential of UDSC as an SD modelling system. We recruited individuals of both sexes. A population of heterogeneous cells were cultured from anonymised urine samples. Stem cells were isolated from the population through FACS and then differentiated into osteogenic or chondrogenic lineage cells using specific media. On day 28 after differentiation induction, the cells showed positive Alizarin Red staining for osteogenesis, indicating mineralised matrix deposition, and positive Alcian Blue staining for chondrogenesis, verifying glycosaminoglycan production. Additionally, qPCR data demonstrate the increase in osteogenic and chondrogenic markers as differentiation progresses.

In conclusion, this project highlights the potential of UDSC as a novel in vitro model for studying SD. In the future, we aim to enhance the utility of this model to facilitate pharmaceutical development and personalised medicine, ultimately improving diagnosis and treatment strategies for rare genetic disorders.

Abstract No. P088

A human iPSC model of tauopathies engineered for 4R Tau isoform expression endogenously develops late-stage neuronal Tau pathology

Angelika Dannert[1,*], *Nathalie Schulz*[1], *Julien Klimmt*[1], *Lea Knez*[1], *Bernhard Groschup*[1], *Carolina Cardoso Gonçalves*[1], *Martina Fetting*[2,3], *Matthias Brendel*[1,4], and *Dominik Paquet*[1,2]

[1] University Hospital, LMU Munich
[2] Munich Cluster for Systems Neurology (SyNergy)
[3] German Center for Neurodegenerative Diseases (DZNE), Munich
[4] SyNergy - Munich Cluster for Systems Neurology, Munich

*Presenting author

Malfunction of the protein Tau is a hallmark of neurodegenerative Tauopathies such as Alzheimer's disease and Frontotemporal Dementia. Molecular mechanisms underlying Tauopathies are still poorly understood, in part due to a lack of human models endogenously developing major disease hallmarks. Adult Tau isoform expression contributes to Tau pathogenesis but is challenging to replicate in human stem-cell-derived systems, which likely impedes formation of late-stage disease phenotypes and hence research on underlying mechanisms and drug development.

We show that induction of adult human brain-like 4R Tau isoform expression in iPSC-derived neurons enables endogenous formation of late-stage Tauopathy hallmarks in iPSC-derived neurons engineered to contain synergistic Tau mutations. Neurons accumulated seeding-competent, hyper-phosphorylated, fibrillar Tau in tangle-like structures. Furthermore, exclusive expression of mutant 4R in the absence of 3R Tau disproportionately intensified pathology, resulting in highly abundant Tau misfolding and aggregation. Finally, we provide proof-of-principle that our model can be translationally applied both to test chemical disease modulators and evaluate a human Tau PET tracer.

Collectively, our model enables novel investigations on endogenous mechanisms of human Tauopathy formation, suggesting a central role of 4R Tau isoform expression for pathogenesis in human neurons. Thereby, our model increases the utility of human iPSC-derived neurons to study adult-onset neurodegenerative diseases. Moreover, it may serve as a platform supporting urgently needed development of disease-modifying drugs.

Abstract No. P089

Impact of CNTNAP2 deficiency on dendritic arborization of human neurons

Saloia Es Sbai[1,*], *Jasmin Aicher*[1], *Luzia Heidrich*[1], *Lydia Fischer*[1], *Martin K. Schwarz*[1,2], and *Oliver Brüstle*[1,2]

[1] University of Bonn Medical Faculty and University Hospital Bonn
[2] LIFE & BRAIN GmbH, Bonn

*Presenting author

Contactin associated protein-like 2 (CNTNAP2), is one of the largest genes in the human genome and highly conserved between different species. CNTNAP2 encodes for the protein CASPR2, a transmembrane protein which is involved in various important processes including synapse formation, migration of interneurons and neuronal arborization. Interestingly, CASPR2 expression is highly upregulated in the human cortex, even compared to other human primates, highlighting the need of a human model to understand its function. Clinically, CNTNAP2 deficiency has been linked to multiple disorders and symptoms including autism spectrum disorder and epilepsy. Previous studies on iPSC-derived CNTNAP2 knockout neurons suggested alterations in neuronal network activity. To explore whether these alterations are due to structural differences, we here set out to assess dendritic arborization in CNTNAP2 ko and isogenic control neurons. Largely excitatory and inhibitory neurons were differentiated from induced pluripotent stem cells (iPSC) using inducible overexpression of NGN2 and DLX2/ASCL1, respectively. To achieve single cells resolution a subpopulation of differentiating cells was transduced with a fluorescence marker using AAVs. Mixed cultures composed of 80% NGN2 and 20% DLX2/ASCL1 neurons were then matured on mouse astrocytes. Morphological reconstruction and Sholl analyses of individual fluorescent neurons revealed an impairment of neurite growth with reduced branching complexity in CNTNAP2-/- neurons, affecting both excitatory and inhibitory subtypes. We are currently employing live imaging to explore whether and to what extent CNTNAP2 deficiency also impacts interneuron migration. Our results obtained so far suggest that CNTNAP2 may have an important role in the structural assembly of neuronal networks.

Abstract No. P090

Inflammation-mediated modulation of synaptic function in SHANK3-deficient human neuronal networks

Christian Felski[1,*], *Martina Quadrubbi*[1], *Michaela Segschneider*[1], *Michael Peitz*[1,2], and *Oliver Brüstle*[1]

[1] Institute of Reconstructive Neurobiology, University of Bonn Medical Faculty
[2] Cell Programming Core Facility, University of Bonn Medical Faculty

*Presenting author

Over recent decades, numerous autism spectrum disorder (ASD) risk genes have been identified, many of which are directly linked to synaptic function and connectivity, positioning them as key targets for understanding ASD-associated pathophysiology. Among these, SHANK3 has emerged as a particularly relevant gene due to its critical role as a postsynaptic density scaffolding protein and its association with Phelan-McDermid syndrome (PMS), a condition frequently accompanied by ASD features. Increasing evidence also implicates neuroinflammation, particularly glial activation, as an additional factor in ASD etiology. An emerging notion is that pro-inflammatory activation of microglia in genetically susceptible individuals may unleash ASD pathophenotypes and contribute to the observed inter-individual phenotypic variability. We set out to investigate this hypothesis using co-cultures of iPSC-derived NGN2-induced neurons (iNs) and microglia. In neuronal monocultures, SHANK3 haploinsufficient iNs exhibit reduced electrophysiological activity relative to wild-type neurons under baseline conditions, while dendritic complexity and synaptic density are unchanged. Neither lipopolysaccharide (LPS) exposure nor microglia-conditioned medium induced genotype-specific effects on neuronal morphology or function. We then established stable, long-term neuron-microglia co-cultures. Presence of microglia resulted in enhanced early network activity in both wild-type and SHANK3+/- neurons, although without genotype-specific effects. Ongoing studies address the question whether inflammatory stimulation of these co-cultures will elicit genotype-specific pathophenotypes in both 2D and 3D cultures.

Abstract No. P091

An innovative human iPSC cardiomyocyte platform to accelerate cardiac drug discovery and target validation

*Benjamin Fischer**, *Hanna Schubert*, *Fatima Kahlid*, *Kamran Honarnejad*, *Shadaan Zulfiqar*, *Caroline Imse*, *Sophie Luellau*, *Ambuj Gore*, *Claudia Ploetzky*, *Anna Carbone*, *Doerthe Fust*, *Sandra Lubitz*, and *Bastian Zimmer*

Evotec SE, Hamburg

*Presenting author

Background

Effective treatment of cardiac diseases requires a deep understanding of disease-relevant cellular mechanisms that drive pathology in the human heart. Traditional cell lines often lack the complexity and genetic context necessary to capture these mechanisms. Human induced pluripotent stem cell (iPSC)-derived cardiomyocytes provide a physiologically relevant and genetically diverse platform to model human cardiac disease biology. Evotec's iPSC platform enables detailed study of disease processes critical for identifying therapeutic targets and developing novel treatments.

Methods & Results

We developed a robust, scalable human iPSC-based cardiomyocyte platform integrating a genetically diverse bank of well-characterized iPSC lines and optimized cardiac differentiation protocols. The platform enables the generation of multiple human-relevant cardiac cell types and model systems, including cardiomyocytes, cardiac fibroblasts, 2D co-cultures of these cell types, 3D cardiac spheroids, and engineered heart tissue (EHT). Genome editing technologies, including CRISPR/Cas9 and RNA interference, are routinely applied at both iPSC and cardiomyocyte stages. These tools enable the introduction of disease-relevant mutations to model specific cardiac pathologies, as well as the validation and exploration of novel target gene functions within a human cellular context.

To assess disease mechanisms and potential target effects, we use multi-electrode arrays (MEA) for electrophysiology, xCELLigence and FLEXcyte for contractility, and calcium transient assays. This enables high-throughput, quantitative, and physiologically relevant phenotyping. Pharmacological testing with well-characterized compounds confirms the platform's sensitivity to cardiac functional changes.

Conclusion

Evotec's iPSC cardiomyocyte platform, combined with the Evotec Heart Atlas, offers a human-relevant system for cardiac disease modeling, drug target validation, and mechanistic insights. Together, these platforms accelerate cardiovascular therapy discovery and increase clinical success through improved human and disease relevance.

Abstract No. P092

Adult ADHD, PARK2 copy number variations and the blood-brain barrier: Metabolic studies in hiPSC-derived in vitro models

*Markus Glaser**, *Laura Ceslik*, *Liane Dieterich*, *Sabine Gätzner*, and *Antje Appelt-Menzel*

University Hospital Würzburg

*Presenting author

Copy number variations (CNVs) in the *PARK2* gene have been shown to be associated with attention deficit hyperactivity disorder (ADHD), potentially through impaired mitochondrial energy metabolism caused by disrupted Parkin-mediated mitochondrial quality control. Compromised blood-brain barrier (BBB) function and elevated oxidative stress are hypothesized to contribute to ADHD pathophysiology by altering the brain's microenvironment. Although stimulant medications can alleviate ADHD symptoms, approximately 40% of adult patients show partial or no response to pharmacological treatment, demonstrating that development of novel medication is necessary.

To investigate *PARK2* CNV-related functional and metabolic changes at the BBB, we established *in vitro* test systems using brain capillary endothelial-like cells (BCECs) derived from a total of 10 different human induced pluripotent stem cell (hiPSC) lines generated from ADHD patients carrying *PARK2* CNVs, and from wildtype individuals with and without ADHD. These models aim to elucidate disease-related mitochondrial dysfunction and serve as a platform for future multi-drug screening efforts.

BCECs from ADHD patients with *PARK2* CNVs show no evidence of impaired barrier function, altered expression of cell-type specific markers, disturbed ATP production, or increased ROS levels. Ongoing analyses include assessments of mitochondrial morphology, mitochondrial membrane potential, and metabolomic profiles. Water-soluble metabolites are analyzed to detect alterations in central metabolic pathways, as their levels can provide insights into mitochondrial dysfunction and associated energy metabolism disturbances.

We aim to deepen understanding of how BBB alterations contribute to ADHD pathology, potentially paving the way for more effective therapies targeting these specific mechanisms. This insight could lead to innovative treatment strategies beyond current pharmacological options.

Abstract No. P093

Minimizing observational bias in cell research: Applying low-impact imaging technologies for reliable datasets

Thomas Gohl[*], *Barbora Burýšková*, *Zuzana Nováková*, and *Peter Zehetmayer*

Telight, Hallbergmoos

*Presenting author

Precise live-cell imaging is essential in cell research, yet the act of observation and sample illumination can influence cell physiology. *In vitro* models—such as 2D cultures, 3D organoids, primary cells, and iPSCs—are influenced not only by intrinsic biology or experimental stimuli, but also by the imaging modalities used. Conventional fluorescence microscopy, while indispensable for molecular and structural studies, can introduce phototoxicity, change proliferation, alter biomass accumulation, and change morphology, particularly during long-term experiments.

Low-impact imaging technologies such as **Quantitative Phase Imaging (QPI)** and **Conical Diffraction Microscopy (CODIM)** help mitigate these effects. QPI provides label-free, quantitative measurement of single-cell dry mass (with the detection sensitivity <1pg) shapes excitation light into localized patterns to minimize phototoxicity and photobleaching, while enabling super-resolution fluorescence imaging.

In sensitive systems, distinguishing genuine biological responses from measurement-induced effects is a persistent challenge. QPI delivers unbiased and quantifiable physiological datasets, preserving native cell behavior for functional analyses. CODIM-based fluorescence imaging provides high-resolution molecular and structural insights with a reduction in light-induced effects compared to conventional fluorescence approaches.

Each method addresses specific needs: QPI excels in long-term, label-free physiological monitoring, whereas CODIM enables minimally invasive fluorescence imaging at high resolution. Both represent strategies for reducing observational bias, safeguarding sample integrity, and improving the accuracy of both functional and molecular analyses in cell research.

Abstract No. P094

Automated high-throughput wound-healing assay for quantitative analysis of cell migration

Reinhild Geisen, *Aleks Guledani**, *Wiebe Schäfer*, *Sebastian Kollenda-Strunk*, *Benjamin Werdelmann*, *Martin Stoer*, *Karsten Schulze*, *Rohan D'Sa*, and *Matthias Pirsch*

SYNENTEC GmbH, Elmshorn

*Presenting author

Cell migration is a fundamental process, playing a key role in tissue development, regeneration, and repair, as well as in pathological processes (Grada et al. 2017). A commonly used in vitro method to study migration is the wound healing assay, in which a cell-free gap (wound) is generated within a confluent cell monolayer and closure is monitored over time by live-cell imaging or repeated microscopy (Jonkman et al. 2014). However, conventional microscopes have limited throughput, allowing only a small number of samples to be analyzed simultaneously.

To overcome this limitation, we developed a high-throughput wound healing assay based on our automation platform comprising CELLAVISTA®, SYBOT X-1000, CYTOMATTM 2 C-LiN, and YT-SOFTWARE®. The automated image analysis algorithm consists of two phases. In the first phase, plates are imaged at 4x magnification, reducing measurement time for a 96 well plate to a minimum of 50 seconds. The YT-SOFTWARE® algorithm automatically identifies and marks cell-free areas as regions of interest (ROIs). In the second phase, these ROIs are re-imaged at 10x magnification, further reducing scanning times down to 37 seconds per 96 well plate.

To validate our wound healing application, we analyzed cell migration of premalignant H6c7-KRAS pancreatic cells. Wounds of varying shape, length, width and orientation were generated by manual scratching (scratch assay) or by silicone inserts (Oris™ and ibidi®). In addition, cells were treated with increasing concentrations of Cytochalasin D, an actin polymerization inhibitor, to impede cell migration. Wound closure was monitored hourly, and the confluence within the gap as well as the average gap width was analyzed and quantified automatically for each treatment condition. The assay successfully detected a dose-dependent inhibition of wound closure by Cytochalasin D.

In summary, this automated high-throughput wound healing assay provides a robust and efficient platform for quantitative analysis of in vitro cell migration.

Grada, A et al. (2017). Research Techniques Made Simple: Analysis of Collective Cell Migration Using the Wound Healing Assay. The Journal of investigative dermatology, 137(2), e11–e16. https://doi.org/10.1016/j.jid.2016.11.020

Jonkman, J E et al. (2014). An introduction to the wound healing assay using live-cell microscopy. Cell adhesion & migration, 8(5), 440–451. https://doi.org/10.4161/cam.36224

Abstract No. P095

Developing antisense oligonucleotide therapies for SCA3 using iPSC-derived neural models

Jacob Helm[1,2], *Luisa Baraban*[1,2], *Melanie Kraft*[1,2], *Yvonne Schelling*[1,2], *Jeannette Hübener-Schmid*[3], *Nicolas Casadei*[3], *Ludger Schöls*[1,2], and *Stefan Hauser*[1,2,*]

[1] German Center for Neurodegenerative Diseases, Tübingen
[2] Hertie Institute for Clinical Brain Research, Tübingen
[3] University of Tübingen

*Presenting author

Background and Objectives: Antisense oligonucleotides (ASOs) are short RNA sequences that bind to complementary (pre-)mRNA in the cytosol or nucleus and alter the processing of the target gene by regulating splicing or initiating degradation through RNase H recruitment. Current methods for developing ASO therapies for neurodegenerative diseases lack a human-relevant and scalable testing platform and rely heavily on animal models. We aim to establish a multimodal platform using iPSC-derived neurons and brain organoids to evaluate ASO efficacy, longevity, specificity, and toxicity using the genetic background of patients in a disease-specific context, focusing on spinocerebellar ataxia type 3 (SCA3).

Methods: We optimised a workflow to identify disease-associated SNPs using a multiplexed, targeted allele-specific sequencing approach. Gapmer ASOs targeting various disease-associated SNPs were screened in iPSC-derived neurons and further validated in cerebellar organoids from SCA3 patients.

Results: Several gapmer ASOs were identified that are able to specifically reduce mutant ataxin-3 protein levels in both iPSC-derived neurons and cerebellar organoids, while maintaining the expression of the wild-type form. Cerebellar organoids will be used for in-depth characterisation of promising ASOs, including acute and long-term toxicity assays and analysis of on- and off-target effects, to enable improved, personalised and patient-centric analysis of ASOs as a treatment for SCA3.

Discussion and Conclusion: iPSC-derived neural models provide a human-relevant platform to evaluate ASOs for neurodegenerative diseases. This platform allows for the assessment of ASO efficacy, off-target effects, acute and long-term toxicity using protein quantification, calcium imaging, NfL levels, and omics technologies. This approach is adaptable to other brain diseases and RNA-based therapeutics.

Abstract No. P096

iPSC-derived cortical brain organoids and neurons for the study of pathomechanisms and gene therapy approaches in Leigh syndrome

Sonja Heiduschka[1,*], *Shattom Ghosh*[2], *Stephanie Le*[1], *Annika Zink*[1], *Caleb Jerred*[1], *Louis Neu*[3], *Jochen Dobner*[4], *Paul Disse*[1], *Nico Melzer*[1], *Christine R. Rose*[3], *Andrea Rossi*[4], *Michal Minczuk*[2], and *Alessandro Prigione*[1]

[1] University Hospital Düsseldorf
[2] University of Cambridge, UK
[3] Heinrich Heine University Düsseldorf
[4] IUF-Leibniz Research Institute for Environmental Medicine, Düsseldorf

*Presenting author

Human induced pluripotent stem cells (iPSCs) and their derivatives highly contribute to the implementation of the 3R (replace, reduce, refine) principle in medical research. Next to their ethical advantages, they can model diseases in which animal models are insufficient, and thus help to overcome the problem of interspecies differences. One of such diseases is Leigh syndrome (LS), a rare mitochondrial disorder with no effective treatment options due to its poorly understood pathomechanism. It manifests in a wide range of especially neurological symptoms caused by mutations in genes responsible for the function of the mitochondrial respiratory chain. New approach methodologies (NAMs), such as iPSC-derived cortical brain organoids (cBO) and neurons, are therefore promising model systems to understand LS pathology.

cBO were generated from LS patient iPSCs with mutations in the mitochondrial gene *MT-ATP6*. They were characterized and compared to control cBO by immunostaining, transcriptomics and microelectrode array. cBO mimicked developmental features of the human brain, such as neurogenic zones and neural marker expression. Patient cBO displayed altered proliferation and pathways related to synapses, as well as reduced electrical spike frequency. Calcium measurements were performed on cultured cortical brain organoid slices (cBOS). Patient cBOS showed an increased calcium response after chemically-induced metabolic stress. To assess potential gene therapy approaches, mitochondrial DNA editing strategies were tested by constructing plasmids containing double-stranded DNA deaminase toxin A-derived cytosine base editors (DdCBE) specific for the correction of two *MT-ATP6* mutations. Patient-derived iPSCs were transfected with DdCBE-containing plasmids, and cBO, cBOS, and iPSC-derived dopaminergic neurons were transduced with adeno-associated virus 9 (AAV9) encoding DdCBE. In ongoing work, the expression of DdCBE is evaluated by fluorescence imaging, PCR, FACS, and Sanger sequencing. In summary, these models show that they can capture disease features and may be used as a platform for setting up gene therapy applications.

Abstract No. P097

Development of iPSC-derived models for succinic semialdehyde dehydrogenase deficiency and their cellular and molecular profiling

Samuel Hofmann[1,*], *Michelle Xi*[1], *The SIMPATHIC Consortium*[2], *Stefan Kölker*[1], and *Sabine Jung-Klawitter*[1]

[1] Heidelberg University, Medical Faculty
[2] Radboud UMC, Nijmegen, The Netherlands

*Presenting author

Succinic semialdehyde dehydrogenase deficiency (SSADHD) is a monogenetic, recessive, rare neurometabolic disorder caused by mutations in the ALDH5A1 gene leading to loss or dysfunction of SSADH, a crucial enzyme in GABA catabolism. SSADHD is characterized by accumulation of GABA and γ-hydroxybutyrate, which is associated with the neurological manifestations of the disease. Patients present with a variety of symptoms including developmental delay, hypotonia, ataxia and epilepsy. Treatment is purely symptomatic and aims at alleviating seizures and psychiatric symptoms, indicating a significant unmet medical need. To understand the molecular pathomechanisms of SSADHD and to facilitate future drug screens, we utilized forward reprogramming to generate glutamatergic neurons from iPSC lines of one SSADHD patient and two carriers, by transduction of a doxycycline inducible expression system for the neurotrophic transcription factor neurogenin 2 (Ngn2).

The generated Ngn2-iPSC lines underwent validation of their iPSC characteristics and were differentiated into glutamatergic neurons. We found mitochondrial dysfunction in SSADHD Ngn2 neurons, as shown by reduction of mitochondrial membrane potential and concomitantly enhanced levels of reactive oxygen species. Additionally, a reduction in overall mitochondrial amount was observed by fluorescence microscopy analysis. The severity of phenotypes correlated with the residual level of SSADH protein as determined by western blot. Taken together, this shows that we can efficiently recapitulate disease-specific phenotypes in our iPSC-based model. Future work includes analysis of neuronal morphology and implementation of additional forward reprogramming protocols for GABAergic neurons using doxycycline inducible expression of Ascl1. All in all, we established a valuable tool for understanding disease physiology and to identify novel therapeutic options in a drug repurposing screen within the scope of the Horizon SIMPATHIC (Accelerating drug repurposing for rare neurological, neurometabolic and neuromuscular disorders by exploiting SIMilarities in clinical and molecular PATHology) consortium.

Abstract No. P098

Rewiring the pre-cancer landscape: KRAS-driven secretome involving TNFα shapes the pancreatic niche

Chantal Allgöwer[1], *Medhanie Mulaw*[2], *Markus Breunig*[1], *Meike Hohwieler*[1,*], and *Alexander Kleger*[1]

[1] Institute of Molecular Oncology and Stem Cell Biology, Ulm University Hospital
[2] Central Unit Single Cell Sequencing, University Ulm

*Presenting author

Background: Pancreatic ductal adenocarcinoma (PDAC) is an aggressive and highly lethal malignancy, dominated by oncogenic KRAS mutations. While the tumor microenvironment (TME) in advanced PDAC has been extensively characterized, the molecular and cellular events that precede invasive transformation remain poorly understood. To address this gap, we investigated early epithelial and microenvironmental changes during the earliest stages of pancreatic tumorigenesis.

Methods: We employed human stem cell-derived pancreatic duct-like organoids (PDLOs) engineered to express mutated KRAS, and performed time-resolved single-cell RNA sequencing and bulk ATAC-sequencing to capture dynamic transcriptional and chromatin changes.

Results: Early $KRAS^{G12D}$ expression elicited robust gene programs associated with extracellular matrix remodeling and inflammation. Machine learning approaches identified transcriptomic classifiers predictive of KRAS-driven niche priming with high accuracy. Various co-culture systems revealed that the $KRAS^{G12D}$-induced secretome activates pancreatic stellate cells (PSCs) and impairs T cell infiltration. In silico reconstruction of a virtual pancreatic cancer landscape mapped interactions between PDLOs and niche components, highlighting TNFα as a top-ranked KRAS-regulated ligand. Functional assays confirmed TNFα's role in both PSC activation and immune evasion.

Conclusion: Our findings uncover critical KRAS-dependent signaling events that prime the tumor microenvironment prior to PDAC onset, offering new insights into the earliest stages of pancreatic cancer development.

Abstract No. P099

Development of autologous iPSC-based immune systems-on-chip for infectious disease research

*Reyk Horland**, *Andy Kleemann*, *Leopold Koenig*, *Thi Phuong Tao*, *Annika Winter*, and *Uwe Marx*

TissUse GmbH, Berlin

*Presenting author

The development of effective medical countermeasures (MCMs) against diverse infectious diseases remains a critical challenge in global health. Here, we introduce a novel strategy employing Immune Microphysiological Systems (Immune Systems-on-Chip) to deepen our understanding of infection biology and improve the assessment of therapeutic interventions.

Our approach centers on enhancing MCM evaluation by modeling key aspects of pathogen-host interactions and immune responses. To this end, we have established a multi-organoid, autologous platform composed of cells derived from a single donor, thereby ensuring physiological relevance and reducing inter-donor variability.

The system integrates three core components: (1) a human lung airway organoid featuring alveolar macrophages (ALMs) to model primary infection sites; (2) a liver organoid to support metabolic function and long-term co-culture stability; and (3) a lymph node organoid that recapitulates immune activation and coordination. These organoids are derived from induced pluripotent stem cells (iPSCs) and donor-derived immune cells.

Utilizing the HUMIMIC Chip platform, the system supports prolonged co-culture and dynamic immune interactions. Initial results demonstrate successful infection of ALMs by model pathogens, triggering activation of innate immune signaling pathways. Co-culture with the liver organoid sustains immune activity and suggests metabolic crosstalk relevant to systemic infection.

The lymph node organoid incorporates monocyte-derived dendritic cells (MoDCs) for antigen presentation, as well as B and T cells to simulate adaptive immune responses. Early findings reveal robust immune activation and antigen-specific T cell responses within the lymphoid microenvironment.

This platform offers a physiologically relevant and scalable tool for accelerating the preclinical evaluation of MCM candidates and improving our understanding of emerging pathogens. Beyond MCM development, the system holds promise for broader applications in infection biology, immunology, and vaccine research—contributing to enhanced preparedness for future infectious disease threats.

Abstract No. P100

A fully human glioblastoma-in-brain-spheroid model enables high-throughput compound screening

Sandra Horschitz[1,*], *Ammar Jabali*[1], *Sophie Heuer*[2], *Eric Zillich*[1], *Lea Zillich*[1], *Dirk Hoffmann*[2], *Akshaya Kumar*[1], *David Hausmann*[2], *Daniel Azorin*[2], *Ling Hai*[2], *Wolfgang Wick*[2], *Frank Winkler*[2], and *Philipp Koch*[1]

[1] Central Institute of Mental Health, Mannheim
[2] German Cancer Research Center, Heidelberg

*Presenting author

In glioblastoma (GBM) the intricate interplay between tumor cells and the surrounding microenvironment plays a crucial role in tumor progression, invasion, and therapeutic resistance. So far, studying these interactions in a controlled and representative model system has been challenging. We have developed hGliCS, a fully human glioma-cortical spheroid model that allows the elucidation of the biology of GBM cells and their interactions with a human-specific brain-like microenvironment and neurons. GBM cells efficiently invade the cortical spheroids, forming a well-connected network of communicating cells. The heterogeneous cellular states of the GBM cells within this model closely resembled findings previously observed in glioblastoma patients and in mouse xenografts. In contrast to the substantial changes observed in the tumor cell population, the impact of the GBM cells on the neurons was minimal.

Further, we evaluated the utility of the hGliCS model in high-throughput screening (HTS) contexts. We conducted a screening campaign using 180 CNS-penetrant small-molecule compounds that are non-classical cytostatic agents. The assay, conducted under miniaturized conditions, proved to be robust and reproducible. Multiple candidate compounds selectively reduced GBM cell viability and connectivity within the spheroid context, highlighting the potential of hGliCS for rapid therapeutic discovery.

These findings highlight the adaptability of the hGliCS model as a powerful translational tool—supporting both mechanistic investigations and scalable drug screening in a fully human, brain-like microenvironment.

Abstract No. P101

Leigh syndrome neurodevelopmental dysfunction modeled in two and three dimensions

Caleb Jerred[1], *Haribaskar Ramachandran*[2], *Andrea Rossi*[2], *Natascia Ventura*[2], and *Alessandro Prigione*[1]

[1] University Hospital Düsseldorf
[2] Leibniz Research Institute for Environmental Medicine, Düsseldorf

Leigh syndrome (LS) is a rare pediatric metabolic disease. Typical diagnostic manifestation includes bilateral lesions in the basal ganglia. Currently, there is no cure and 50% of patients may die by the age of three. Modeling the disease has remained challenging due to the lack and inadequacy of animal models. Furthermore, there are >100 of monogenic origins, of which mitochondrial complex I deficiencies are the most common. Stem cell models offer an opportunity to study the unique human pathology of the disease in a controlled genetic context. Here, we use a mutant cell line harboring a Leigh syndrome mutation in mitochondrial complex I component NDUFS1, paired with an isogenic control. Neurodevelopmental pathology was investigated using neural progenitor cells (NPCs). Midbrain neurodegenerative pathology was explored in 2- and 3-dimensions using midbrain dopaminergic neurons (mDANs) and midbrain organoids (MBOs).

Mutant NPCs demonstrated a reduced mitochondrial membrane potential and an early branching phenotype. Mutant mDANs displayed atypical morphology, a reduction in axonal branching, and a reduced mean firing rate in the most active neurons. Mutant MBOs showed reduced growth rate and increased mean firing rate under epileptic stimulation. Together, these data demonstrate that *NDUFS1* mutation disturbs neurodevelopment and causes structural and functional changes in midbrain models. Additionally, they suggest that LS disease pathology such as epilepsy is represented in the 3D systems. Furthermore, they demonstrate the benefit of combining 2- and 3-dimensional modeling to capture both cellular and systemic dysfunction. These models and phenotypes will be beneficial in understanding mechanistic disease progression and identifying treatment options.

Abstract No. P102

Development of a high-throughput screening method for drug target discovery in schizophrenia by means of utilizing co-cultured hiPSC-derived cortical neurons and microglia

Jessica Jung[1,2,*], *Julia Wangemann*[1], *Sandra Horschitz*[1,3], *Anne Hoffrichter*[1,2], and *Philipp Koch*[1]

[1] Central Institute of Mental Health (ZI), Mannheim
[2] Heidelberg University/Medical Faculty Mannheim
[3] German Cancer Research Center (DKFZ), Heidelberg

*Presenting author

Schizophrenia is a highly heritable psychiatric disorder affecting about 1% of the global population, characterized by grey matter loss, impaired network connectivity, and reduced synapse density. Genetic studies highlight the complement component 4 (C4) locus as a major risk factor, with higher C4A activity correlating with excessive synaptic pruning, most consistently observed in the context of microglial involvement.

To investigate this mechanism, we employ a human in vitro assay based on co-cultures of hiPSC-derived cortical neurons and microglia. Overexpression of C4A in neurons enhances microglial activation and synapse engulfment, leading to pronounced synaptic deficits, while inhibition of the microglial NLRP3 inflammasome with oridonin restores synaptic density, pointing to a potential therapeutic strategy.

Building on this foundation, we are advancing the assay into a high-throughput compatible format using the *Revvity Opera Phenix Plus* High-Content Screening system combined with the *explorer G3 workstation*. A key step in this transition has been the implementation of a new guided differentiation protocol aimed at reducing experimental time through the rapid and reliable generation of homogeneous cortical neuronal cultures. Ongoing work also includes the development of a cryopreservation protocol for microglia to establish a cell bank that facilitates rapid and reproducible large-scale applications. In parallel, an automated analysis pipeline has been developed within the *Harmony* system for quantitative assessment of synaptic pruning, enabling efficient analysis of large-scale datasets. The pipeline also incorporates automated detection and classification of neuronal and microglial nuclei without the need for a microglial marker, thereby enhancing assay robustness and efficiency. Together, these improvements enable systematic and scalable testing of candidate compounds, providing a powerful platform for discovering potential drugs.

Abstract No. P103

GTP cyclohydrolase I deficiency leads to mitochondrial impairment in iPSC-derived neural progenitor cells and Ngn2-induced neurons

Sabine Jung-Klawitter[1,*], *Samuel Hofmann*[1], *Luciana Hannibal*[2], *The SIMPATHIC Consortium*[3], *Georg F. Hoffmann*[1], *Thomas Opladen*[1], and *Daniel Hübschmann*[4]

[1] Heidelberg University, Medical Faculty Heidelberg
[2] Medical Center University Freiburg
[3] EU Consortium, Radboud UMC, Nijmegen, The Netherlands
[4] German Cancer Research Center (DKFZ), Heidelberg

*Presenting author

GTP cyclohydrolase I (GTPCH) is the first and rate-limiting enzyme in tetrahydrobiopterin (BH_4) biosynthesis, producing an essential cofactor for aromatic amino acid decarboxylase and nitric oxide synthases (NOS). Low levels of BH_4 associated with GTPCH deficiency (GTPCHD) affect neurotransmitter biosynthesis and can cause uncoupling of NO-synthesis. By using patient-specific induced pluripotent stem cells (iPSCs) we investigated the pathological features of GTPCHD and how the deficiency affects the cellular metabolism of neural progenitor cells (NPCs).

Within bulk RNA Sequencing data of GTPCH-deficient NPCs, via gene set enrichment analyses we identified dysregulation in neuronal differentiation and maturation. Another pathway affected by GTPCHD in NPCs involved oxidative phosphorylation and mitochondrial structural proteins. We investigated the respiratory capacity of GTPCH-deficient NPCs via Seahorse assays and measured TCA cycle metabolites. Enhanced oxygen consumption rate of the deficient cells and subtle changes in TCA cycle metabolites were found. Moreover, electron microscopy images of GTPCH-deficient NPCs show structural changes in mitochondrial morphology. Deficient cells also show a mild decrease in mitochondrial membrane potential as detected by TMRM staining and enhanced levels of reactive oxygen species. Additionally, axonal and dendritic outgrowth is impaired in GTPCH-deficient Ngn2-induced glutamatergic neurons. Taken together, GTPCH-deficient neuronal cell types show impaired mitochondrial function and morphological changes also found in neurodegenerative diseases. Our next steps will focus on the identification of the functional link between BH_4 deficiency and mitochondrial impairment. Additionally, these phenotypes will be used within the scope of the EU-funded SIMPATHIC (Accelerating drug repurposing for rare neurological, neurometabolic and neuromuscular disorders by exploiting SIMilarities in clinical and molecular PATHology) consortium to identify novel therapeutic targets based on a drug repurposing screen.

Abstract No. P104

Beyond plastic: Engineering soft environments with ultra-high viscosity alginate for hiPSC-derived neurons in vitro

Isabell Karnatz[1,2,*], *Beate-Sophie Nenninger*[1,2], *Jeanette Wihan*[2], *Michael Gepp*[2], *Heiko Zimmermann*[1,2], and *Julia Neubauer*[2]

[1] Saarland University, Saarbrücken
[2] Fraunhofer Institute for Biomedical Engineering IBMT, Sulzbach

*Presenting author

The human brain is one of the softest tissues in the body, characterized by an exceptionally low stiffness in the sub-kilopascal range. Mimicking both, the cellular composition and the mechanical property of native tissue is crucial to enhance the transferability of *in vitro* findings to human physiology and disease. Although neurons are known to be mechanosensitive, standard culture systems still rely on rigid plastic surfaces that poorly reflect the brain's native biomechanical environment. To overcome this issue, we investigated the use of ultra-high viscosity (UHV) alginates as an inert and adjustable hydrogel platform for neuronal cell culture. UHV-alginate allows a precise tuning of biophysical properties such as stiffness and viscoelasticity. It can also be modified with extracellular matrix proteins, e.g. laminin, to support neuronal cell adhesion. In this study we developed a 2D culture workflow using UHV-alginate as substrates for human induced pluripotent stem cell (hiPSC) derived NGN2 neurons. Nanoindentation measurements of UHV-alginate substrates revealed stiffness values below 1 kPa, which are within the physiological range of cortical tissue. Subsequently, the maturation of NGN2 neurons on hydrogels with varying Young's moduli was compared to conventional plastic cultureware. AI-assisted morphological analysis were employed to evaluate neuronal development and revealed distinct neurite-network patterns on hydrogel substrates compared with plastic. In addition, neuronal marker gene expression was quantified. For calcium-imaging, NGN2 neurons were co-cultured with astrocytes to assess network activity. Our results identify UHV-alginate as a tunable, calcium-imaging-compatible platform, potentially addressing the limitations of conventional rigid plastic and suitable for compound screening. Taken together, combining hiPSC-derived neural models with soft hydrogel substrates move us closer to achieving *in vivo*-like neuroscience in a dish.

Gepp, M. M. et al. Bioactive surfaces from seaweed-derived alginates for the cultivation of human stem cells. J. Appl. Phycol. 29, 2451–2461 (2017)

Wihan, J. et al. Production of Human Neurogenin 2-Inducible Neurons in a Three-Dimensional Suspension Bioreactor. J. Vis. Exp. (2023)

Abstract No. P105

Modelling PURA syndrome in cerebral organoids uncovers cell type misdistribution as likely cause for symptoms in patients

Carolin Ketteler[1,*], *Sabrina Bacher*[1], *Silvia Marcato*[1], *Ejona Rusha*[2], *Anne Dück*[3], and *Dierk Niessing*[1, 4]

[1] Institute of Structural Biology, Helmholtz Munich
[2] Institute of Stem Cell Research, Helmholtz Munich
[3] Institute of Pharmacology and Toxicology, Helmholtz Munich
[4] Institute of Pharmaceutical Biotechnology, Helmholtz Munich

***Presenting author**

The neurodevelopmental disorder PURA syndrome is caused by heterozygous *de novo* mutations in the *PURA* gene. The disorder is characterized by various symptoms including intellectual disability, hypotonia and epilepsy. Previous PURA studies have mainly focused on HeLa cells and rodent neuronal cells, as well as different mouse models, which showed partially contradictory results. However, models accurately reflecting human neurodevelopment remain lacking.

To address this gap, we generated human homozygous knock-out (KO) iPS cell lines and patient-derived iPS cell lines harboring the most recurrent pathogenic PURA mutation (Phe233del), alongside their respective isogenic controls. These cells were differentiated into neural progenitor cells (NPCs) and cerebral organoids, followed by transcriptome profiling and single-cell RNA sequencing.

Morphological and size analysis of NPCs and organoids revealed no difference between KO, patient, and their isogenic control cell lines. However, transcriptomic analysis upon PURA KO identified misregulation of specific RNAs critical for NPC maintenance and cell fate decisions during early neuronal differentiation. Notably, in 3D organoids, these molecular alterations result in a decrease of GABAergic neurons and an overproduction of glutamatergic neurons, as well as non-neuronal cortical hem and choroid plexus cells.

Our findings highlight a critical role of PURA in early embryonic development and suggest that the neurodevelopmental phenotypes observed in patients may arise from a misdistribution of cell types during early brain development.

Abstract No. P106

Advancing and modulating human heart-forming organoids (HFOs) for drug testing

Felix Kleemiß[1,*], *Lars Leonhardt*[1], *Liam Wilson*[1], *Miriana Dardano*[1], *Jana Teske*[1], *Annika Franke*[2], *Lika Drakhlis*[1], and *Robert Zweigerdt*[1]

[1] Hannover Medical School (MHH)
[2] Lebao, MHH

*Presenting author

Although their numbers were drastically decreased since the 1970s, animal models still play a major role in life sciences and drug development, despite having numerous drawbacks such as high costs and low predictive capabilities for the human organism. Therefore, organoids derived from human pluripotent stem cells (hPSCs) play a key role in the replacement of animal models in disease modelling and drug screening. The heart-forming organoid (HFO) system is a hPSC-derived pre-vascularized and patterned multi-tissue organoid model that closely resembles early heart and foregut development [1]. To establish HFOs as a platform for drug screening, we exposed them to FDA-approved antiangiogenic chemotherapeutic compounds. Their effect on the vascular network and other tissue types within the HFO was evaluated by flow cytometry (FC) and whole-mount immunofluorescence staining (WMS). Confocal images were processed using Python-script and analysed using AngioTool, which allowed a quantitative analysis and characterisation of the vascular network. Treating HFOs with Axitinib and Cabozantinib in a clinically relevant concentration range (cmax) resulted in a strong antiangiogenic effect without decreasing the viability of the organoids. Both compounds did not show any effect on the cardiac and pluripotent compartment of the HFO while altering the expression pattern of endodermal surface markers. These preliminary results highlight the HFOs' potential as a robust drug screening platform to test antiangiogenic compounds while simultaneously investigate possible off-target effects on cardiac and foregut tissues.

Drakhlis, L., Buswanath, S., Farr, CM., Zweigerdt, R. et al. (2021). Nat Biotechnol 39,737-746. doi:10.1038/s41596-021-00629-8

Abstract No. P107

ALS causative KIF5A mutations lead to translational impairment and nucleolar stress in hiPSCs derived motor neurons

Rosanna Parlato[1], *Rüstem Yilmaz*[1], *Francesca Tuorto*[1], *Babak Loghmani*[1], *Philipp Koch*[1,2,*], *Jochen Weishaupt*[1,3], *Elena Cairo*[1], and *Johannes Wilbertz*[4]

[1] Universität Heidelberg
[2] Central Institue of Mental Health, Mannheim
[3] Universität Ulm
[4] Ksilink, Strasbourg, France

*Presenting author

Our group has previously described heterozygous ALS-causing mutations in the Kinesin Family Member 5A (KIF5A). KIF5A is a kinesin responsible for anterograde transport of proteins, organelles, RNA, and neurofilaments along the neurites. ALS-linked mutations occur in the kinesin's C-terminal cargo-binding domain and are caused by the disruption of exon 27 splicing. Indeed, a mutation resulting in KIF5A exon 27 skipping (ΔExon27) has been shown to cause altered protein and RNA interactions. Moreover, we recently found that the conformational changes caused by ΔExon27 is inducing neurotoxicity by abolishing KIF5A autoinhibition. We generated hiPSCs from one pre-manifest carrier of a heterozygous ΔExon27 mutation (c.3020+2T>C) and from two ALS patients of a family with a c.2993-1G>A heterozygous mutation (KIF5A-ALS). These mutations alter exon 27 splicing differently, yet both are predicted to lead to the production of a common C-terminal aberrant end. In addition, patient-derived motor neurons displayed a significant increase in KIF5A inclusions in comparison to control.

Interaction analysis in HEK293T cells revealed altered binding of mutant KIF5A, for example to proteasome subunits, deubiquitinases, and ribosomal components. We validated these findings in the KIF5A-ALS motor neurons using proximity ligation assays. Notably, we observed increased expression of ribosomal proteins and a decreased global translation, suggesting an early compensatory response to impaired ribosome biogenesis. Furthermore, bulk RNA sequencing of motor neurons at differentiation days 20, 35, and 50 from four control and four KIF5A-ALS lines revealed maturation-dependent dysregulation of pathways related to neuronal structure and protein synthesis. Collectively, these results reveal a novel pathomechanism dependent on KIF5A-ALS mutations for the discovery of therapeutic targets modulating disease progression.

Abstract No. P108

Identification and mitigation of major confounders in iPSC-based psychiatric disease modeling

Tamara Krutenko[1,], Karola Hünten[2], Christian Felski[1], Yevheniia Kyriachenko[1], Amael Kerber[1], Friederike David[2], Eva Beins[2], Sugirtahn Sivalingam[3], Bettina Burger[4], Lea Sirignano[5], Fabian Streit[5], Frederike Stein[6], Petraina Pfefferle[7], Stefan Herms[2], Per Hoffmann[2], Tilo Kircher[6], Udo Dannlowski[8], Marcella Rietschel[5], Michael Peitz[1], Sven Cichon[4], Markus Nöthen[2], Andreas Forstner[2]*, and *Oliver Brüstle[1]*

[1] Institute of Reconstructive Neurobiology, University of Bonn Medical
[2] Institute of Human Genetics, University of Bonn, School of Medicine
[3] Core Unit for Bioinformatics Data Analysis, Medical Faculty, University of Bonn
[4] Department of Biomedicine, University of Basel, Switzerland
[5] Department of Genetic Epidemiology in Psychiatry, Central Institute of Mental Health, Medical Faculty Mannheim, University of Heidelberg
[6] University Hospital for Psychiatry and Psychotherapy, UKGM, Philipps-Universität Marburg
[7] Medical Faculty, Comprehensive Biobank Marburg (CBBMR), Philipps University of Marburg
[8] Institute for Translational Psychiatry, University of Münster

*Presenting author

While iPSC-based models provide a powerful tool for studying psychiatric disorders, even small variabilities related to interclonal homogeneity, robustness of differentiation protocols, and standardization of supplies can compromise reproducibility and data interpretation. Using risk-stratified patient-derived iPSCs and a genome editing-based candidate gene approach, we scrutinized key obstacles impacting iPSC-based modeling of depression-associated neurodevelopmental disorders. Using a cohort of patients and healthy donors, we generated cortical neural progenitors stratified by polygenic risk scores for bipolar disorder (BD1) and assessed neurodevelopmental phenotypes. We also employed a genome editing approach to study GPM6A, a gene linked to depression, schizophrenia, and BD1. Confounders, including clonal variability, donor blood mosaicism, and culture condition,s were systematically evaluated. In the BD1 model, clonal variability largely impacted reproducibility. Neurodevelopmental phenotypes were further highly dependent on iPSC handling and subtle differences in differentiation dynamics. Furthermore, network activity in GPM6A-deficient cortical neurons was biased by cell density and clone-specific culture composition. After addressing these confounders, only few of the previously reported pathophenotypes related to BD1 and GPM6A could be confirmed. However, we identified several new interesting BD1-associated candidates, including NBPF3, a gene involved in the regulation of vitamin B6 clearance and previously linked to symptom severity in schizophrenia. Our findings underscore the critical need for improved standardization in iPSC-based models, especially for polygenic disease models and candidate genes with small effect sizes. Patient-derived models require extended cohorts, optimized donor selection, and advanced standardized protocols incorporating genetic and environmental risk factors. In isogenic models, expanded genetic backgrounds and robust culture systems are essential. Both lines of research depend on careful PSC clone selection, standardized supply of high-end materials, and operator consistency. Such advancements are also vital for enhancing the reliability of iPSC models in therapy development and compound testing.

Abstract No. P109

Kidney organoids as a human iPSC-derived in vitro model for Fabry nephropathy

Sandra D. Laufer[1,], Anna Reinelt[1], Götz Rövenstrunk[1], Milagros Wong[1,2], Fabian Haas[1], Linda Blomberg[3], Christine E. Kurschat[3], Nicola Wanner[1], Nicole Muschol[1], Annika Gathmann[1], Nikolay Zolotarev[1], Victor G. Puelles[1,2], Arne Hansen[1], Tobias B. Huber[1]*, and *Fabian Braun[1]*

[1] University Medical Center Hamburg-Eppendorf
[2] Aarhus University
[3] Center for Molecular Medicine Cologne and Center for Rare Diseases, University of Cologne

*Presenting author

Background: Currently, the renal phenotype of Fabry disease cannot be accurately modelled in rodent systems. Our project therefore aims to establish an advanced human *in vitro* system to enhance our molecular understanding of Fabry nephropathy. To facilitate this, we utilize hiPSCs derived from Fabry patients and differentiate them into kidney organoids.

Methods: Primary urinary cells (PUCs) collected from Fabry patients with various pathogenic variants were reprogrammed into hiPSCs and CRISPR/Cas9 gene editing was used to establish isogenic controls. Following a published protocol, we differentiated these cell lines into kidney organoids.

Results: We confirmed the loss of alpha-galactosidase A protein, deficiency of enzyme activity and the accumulation of globotriaosylceramide (Gb3) in patient-derived hiPSCs and kidney organoids. Additionally, targeted metabolomic analysis confirmed the presence of Gb3 subspecies in both systems and elevated LAMP1 staining intensity in immunohistochemistry of diseased kidney organoids suggested an increase in lysosomal mass. This pathophysiological phenotype did not affect differentiation properties, ensuring that our kidney organoids contained nephron specific cell types, shown in snRNA-seq data and immunofluorescence. We implemented treatment with enzyme replacement therapy and, when applicable, chaperone therapy. In both the hiPSCs and kidney organoids, the treatment resulted in a significant increase in enzyme activity and a consequent reduction in substrate accumulation. Furthermore, we investigated the involvement of the innate immune system, specifically the local production and activation of complement components.

Conclusion: We have established a novel and unique discovery platform for human Fabry nephropathy. Ongoing experiments aim to investigate the published pathological accumulation of synuclein alpha in Fabry nephropathy in our *in vitro* system. Current analyses are focused on identifying differential gene expression from the snRNA-seq dataset which will then be complemented with deep proteomic and spatial phenotyping combined with the administration of available therapies.

Support: Amicus Therapeutics, Federal Ministry of Research, Technology and Space

Abstract No. P110

Advanced functional assays reveal co-culture and substrate-driven maturation and patient-specific differences in hiPSC cardiomyocytes and neurons

Marta Lemme[1,], Nina Brinkwirth[1], Nadine Becker[1], Julia Neubauer[2], Isabell Karnatz[2], Jeanette Wihan[2], Bettina Lickiss[3], Peter Linder[3], Ulrich Thomas[1], Ravi Vaidyanathan[4], Simon Hilcove[4], Stuart Prime[5], Matthias Gossmann[3], Elena Dragicevic[1], Alison Obergrussberger[1], Sonja Stoelzle-Feix[1]*, and *Niels Fertig[1]*

[1] Nanion Technologies GmbH, Munich
[2] Fraunhofer-Institute for Biomed. Engin. (IBMT), Sulzbach
[3] innoVitro GmbH, Jülich
[4] FUJIFILM Cellular Dynamics Inc., Tokyo, Japan
[5] Axol Bioscience, Babraham, UK

*Presenting author

Human induced pluripotent stem cell (hiPSC)-derived neurons and cardiomyocytes provide high human relevance for preclinical research while avoiding ethical concerns of animal use. However, their immature phenotype in standard culture limits functional predictivity. Here, we apply complementary biophysical and co-culture strategies to enhance maturation and enable disease-specific discrimination.

The FLEXcyte 96 platform featuring ultra-thin polydimethylsiloxane (PDMS) membranes with in vivo-like stiffness, produced sustained increases in contractile force and reduced beat rate in cardiomyocytes, consistent with an adult-like phenotype, in 2D and in 3D. Gene profiling revealed upregulation of MYH7, ACTC1, and ACTN2, alongside downregulation of fetal genes such as MYH6 and HCN4. Pharmacological testing showed expected adult-like responses to ionotropic compounds such as isoproterenol, S-BayK, and omecamtiv mecarbil. Additionally, Geltrex-coated wells containing co-cultures of 100,000 hiPSC-cardiomyocytes and 10,000 fibroblasts exhibited enhanced maturation, as evidenced by increased contractile force.

High throughput automated patch clamp (APC) was used to record hiPSC-neurons with excellent success rates. Using the SyncroPatch 384 we showed that neuron-astrocyte co-culture increased NaV current expression from 59% to 100% and KV currents from 57% to 85%, with higher amplitudes. Ligand-gated receptor profiling confirmed responses to acetylcholine, GABA, glycine, and glutamate. In another set of experiments, hiPSC-derived motor neurons from healthy controls were compared with ALS C9orf72 variants. Differences in KV current amplitudes and kinetics, along with changes in NaV current amplitudes were observed. Responses to glutamate, glycine and GABA were also detected, with moderate differences in GABA-mediated responses observed in the C9orf72 variants compared with control.

These findings highlight the importance of integrating physiologically relevant mechanical environments with targeted co-culture strategies, coupled to high-throughput functional recordings. Such pro-mature cell models yield hiPSC-derived cardiomyocytes and neurons with enhanced structural, genetic, and electrophysiological maturity, with strong potential for improving the translational accuracy of disease modelling, safety pharmacology, and drug discovery.

Abstract No. P111

A novel KIT D816V mutated hiPSC-mast cell model with IgE-dependent activation

Yanyan Luo[1,2,*], *Jiajun He*[1,2], *Valeria Fernandez Vallone*[3], *Duncan C.Miller*[1,2], *Mengjie Hu*[1,2], *Eric Blanc*[4], *Stefan Frischbutter*[1,2], *Harald Stachelscheid*[3], *Dieter Beule*[4], *Jörg Scheffel*[1,2], and *Frank Siebenhaar*[1,2]

[1] Institute of Allergology, Charité - Universitätsmedizin Berlin
[2] Fraunhofer Institute for Translational Medicine and Pharmacology ITMP, Immunology and Allergology IA, Frankfurt a. Main
[3] BIH Core Unit pluripotent Stem Cells and Organoids, Berlin Institute of Health, Charité - Universitätsmedizin Berlin
[4] BIH CUBI - Core Unit Bioinformatics, Berlin Institute of Health, Charité - Universitätsmedizin Berlin

*Presenting author

Introduction
Mastocytosis is a disorder caused by an excess number of mast cells (MCs) gathering in tissues. The most frequent clinical subtype is indolent systemic mastocytosis (ISM), which is commonly associated with an acquired somatic gain-of-function KIT-D816V mutation. There are currently only limited MC models available, such as HMC1.2, HMC1.3 and ROSA MC$^{KIT-D816V}$. However, they exhibit limitations due to complex genetic backgrounds and the lack of activation, hindering the precise study of specific KIT-D816V-driven pathogenesis.

Method
We have generated a new KIT-D816V mutated induced pluripotent stem cell derived mast cell model (hiPSC-MC$^{KIT-D816V}$) via CRISPR/Cas9 gene editing. To characterize the new cell model, we performed whole genome sequencing and assessed the immunophenotype, survival, proliferation, degranulation and the response to various tyrosine kinase inhibitors (TKIs).

Results
hiPSC-MCs$^{KIT-D816V}$ were rigorously confirmed to carry only the KIT-D816V mutation, and so to be free from other to date described disease-associated mutations. hiPSC-MCs$^{KIT-D816V}$ express typical MC surface markers and degranulate in response to IgE/anti-IgE. In addition, the hiPSC-MCKIT-D816V exhibits auto-phosphorylation of KIT, SCF-independent survival and imatinib resistance, mirroring key features of ISM.

Conclusions
The novel hiPSC-MCKIT-D816V is a functional MC model for preclinical research on mastocytosis.

Abstract No. P112

Modeling PURA syndrome: impact of PURA knockout and F233del mutation on neural progenitor cells

Sabrina Bacher[1], *Silvia Marcato*[1,*], *Carolin Ketteler*[1], *Ejona Rusha*[1], *Micha Drukker*[2], and *Dierk Niessing*[1,3]

[1] Helmholtz Munich
[2] Leiden University, The Netherlands
[3] Ulm University

*Presenting author

Heterozygous *de novo* mutations in the PURA gene cause the neurodevelopmental disorder PURA syndrome. This disease is characterized by mild to severe intellectual disability, hypotonia, epilepsy, and other symptoms, which vary considerably among the more than 700 reported patients. PURA encodes an RNA-binding protein that regulates key cellular processes, including immune response, processing (P-) bodies, and mitochondrial function. However, PURA's role in neurodevelopment and its contribution to PURA syndrome remain poorly understood.

To address this, we modeled the disease in vitro using homozygous knock-out (KO) iPSC lines and patient-derived iPSCs carrying the most recurrent PURA syndrome-causing mutation (F233del). These iPSCs were differentiated into neural progenitor cells (NPCs) to investigate the effects of PURA loss and F233del mutation during early neurodevelopment. While no morphological differences were observed between KO, patient, and isogenic control NPCs, transcriptome profiling identified a set of PURA-regulated RNAs involved in critical processes of early neural development.

Abstract No. P113

Human 3D neuromuscular organoids to model TDP-43 related mechanisms in ALS

Aylin Nebol[*], *Adrianos Skaros*, *Ismail Amr El-Shimy*, *Sara De Vincentiis*, *Iacopo Bicci*, *Inês Afonso Martins*, and *Mina Gouti*

Stem Cell Modeling of Development and Disease Group, Max Delbrück Center, Berlin

*Presenting author

Amyotrophic lateral sclerosis (ALS) is a progressive neurodegenerative disease characterized by motor neuron degeneration, skeletal muscle atrophy, and paralysis. While traditional monoculture systems and assembled co-cultures of neurons and muscles have provided valuable insights into ALS biology, they fail to capture the early cross-tissue interactions and bidirectional signaling at the neuromuscular junctions (NMJs) that are critical for physiological relevance. To address this limitation, we established self-organized neuromuscular models that recapitulate key embryonic developmental principles, offering a major advantage for studying human neuromuscular biology and disease pathophysiology. We established 3D neuromuscular organoids (NMOs) (Martins et al, 2020) and 2D self-organizing neuromuscular junctions (Urzi et al, 2023) from induced pluripotent stem cells (iPSCs) via bipotent neuromesodermal progenitors (NMPs). These systems self-organize into spinal cord neurons and skeletal muscle and form functional NMJs, enabling detailed analysis of ALS-related phenotypes.

Here, we generated NMOs from isogenic iPSC lines carrying heterozygous and homozygous TDP-43^{M337V} mutations alongside the parental KOLF2.1J line to investigate disease mechanisms. By integrating automated high-content image analysis, functional assays, and bulk and single-cell molecular readouts, we map the temporal and spatial relationships between TDP-43 mis-localization, motor neuron vulnerability, and muscle maturation. Collectively, our findings demonstrate that NMOs reproduce key aspects of ALS pathology in the absence of external stress. This establishes NMOs as a developmentally accurate and scalable human platform for the mechanistic dissection of ALS and for preclinical evaluation of therapeutic strategies.

Martins, J.M., Fischer, C., Urzi, A., ..., Gouti, M., Self-Organizing 3D Human Trunk Neuromuscular Organoids, Cell Stem Cell, 2020

Urzi, A., Lahmann, I., Nguyen, L.V.N., ..., Gouti, M., Efficient generation of a self-organizing neuromuscular junction model from human pluripotent stem cells. Nature Communication, 2023

Abstract No. P114

Repeated rhinovirus infections drive epigenetic memory and senescence

Wasifa Nurieva[1,2,*], *Sneha Nemani*[1,2], *Marua Abu Risha*[3], *Uta Jappe*[3], *Janette K. Burgess*[4], *Silke Meiners*[5], *Karosham Diren Reddy*[1,6], *Shauni B. Biedermann*[1,2], *Antonia Bohnhorst*[1,2], *Gesine Hansen*[7,8], *Erika von Mutius*[9,10], *Klaus F. Rabe*[2,11], *Anna-Maria Dittrich*[7,8], *Bianca Schaub*[10,12], *Christine Happle*[7,8], *Lea L. Kronziel*[13], *Inke König*[13,2], *Matthias Kopp*[14], *M. A. Alejandre Alcázar*[15,16], *Folke Brinkmann*[1,2], and *Markus Weckmann*[1,2]

[1] UKSH, Div. Pediatric Pneumology & Allergology, Lübeck; [2] Member of the German Center for Lung Research (DZL), Airway Res. Center North, ARCN, Borstel, Lübeck, Kiel, Grosshansdorf; [3] Res. Center Borstel Leibniz Lung Center, Clinical & Mol. Allergology, Borstel; [4] Faculty Medical Sciences/UMCG, Groningen Res. Inst. Asthma & COPD (GRIAC), Groningen, The Netherlands; [5] Immunology and Cell Biology, Res. Center Borstel Leibniz Lung Center; [6] Epigenetic of Chronic Respiratory Diseases, Res. Center Borstel; [7] Dep. of Pediatric Pneumology, Allergology and Neonatology, Hannover Medical School (MHH); [8] Biomed. Res. End-stage and Obstructive Lung Dis. Hannover (BREATH), DZL Member; [9] German Res. Center Environmental Health (CPC-M), Munich, DZL Member; [10] Dr. von Hauner Children's Hospital, Dep. Pediatrics, LMU University Hospital, Munich; [11] Dep. of Pneumology, LungenClinic Grosshansdorf; [12] Comprehensive Pneumology Center (CPC-M), LMU Munich, DZL Member; [13] Uni. Lübeck, Inst. Med. Biometry & Statistics; [14] Dep. of Pediatric Respiratory Medicine, Uni. Children's Hosp. Bern, Switzerland; [15] Transl. Exp. Pediatrics – Exp. Pulmonology, Uni. Cologne; [16] Dep. Pediatric & Adolescent Med., Excellence Cluster on Stress Reponses in Aging-associated Diseases (CECAD) and (CMMC), Uni Hospital Cologne

*Presenting author

Background: Recurrent human rhinovirus (HRV) infections are linked to childhood asthma development, potentially inducing trained innate immunity (TRIM) via epigenetic changes. Epigenetic aging (DNAge) clocks indicate increased aging rates with asthma onset. Our previous data indicated loss of epithelial stemness and premature aging by repeated HRV infections, questioning the relationship between epithelial TRIM, stemness, and aging.

Objective: To assess how recurrent HRV infections impact stemness loss and aging, distinct from TRIM.

Methods: Bronchial epithelial cells (BEAS-2B) were repeatedly infected with HRV16. After 24 hours, cells were split for DNA/RNA extraction and reseeded for up to five repeated infections. Nasal epithelial cells (NECs) from asthmatic children of the ALLIANCE cohort were analysed after HRV16 infection (DNA methylation, RNA-seq).

Results: In BEAS-2B, the early progenitor marker TP63 decreased ($p<0.01$), while basal epithelial progenitor (BEP) markers KRT5 and LAMA3 increased ($p<0.0.5/0.01$) with infection number. Cluster analysis identified four groups, with cluster 4 (high BEP expression) being enriched in highly infected cells (Chi^2 $p<0.01$). DNAge (HorvathSkin clock) increased with infection number ($p<0.01$), while pro-inflammatoryproteins (e.g. IL-8, IL-1b, both $p<0.01$) declined. In NECs, clusters with higher stemness had lower senescence marker expression ($p<0.001$). DNAge trended with clustered expression markers of senescence.

Conclusions: Repeated HRV infections induced TRIM and senescence. TRIM predominated in cells with low DNAge, while senescence and stemness loss dominated with repeated infections. NECs confirmed higher senescence with reduced stemness. Further studies are needed to clarify the interplay between TRIM, aging and early origins of lung disease.

Abstract No. P115

Patient iPSC-derived microglia reveal disease-associated changes in BPAN

Gamze Özata[1,*], *Rachel Wise*[1], *Aida Cardona-Alberich*[1], *Naiyareen F. Mayeen*[1], *Stephan A. Müller*[2,3], *Stefan F. Lichtenthaler*[2,3], *Luigi Zecca*[4], and *Lena F. Burbulla*[1,3]

[1] Ludwig Maximilian University of Munich
[2] Technical University of Munich
[3] German Center for Neurodegenerative Diseases (DZNE), Munich
[4] Institute of Biomedical Technologies, National Research Council of Italy, Segrate (Milan), Italy

*Presenting author

Beta-propeller protein-associated neurodegeneration (BPAN) is a rare, X-linked neurodegenerative disorder caused by mutations in the *WDR45* gene, leading to impaired autophagy, progressive cognitive decline, seizures, and movement abnormalities from early childhood, with iron accumulation and parkinsonism manifesting in adulthood [1]. Neuroinflammation is increasingly recognized as a key driver of disease progression, yet the underlying cellular mechanisms remain poorly defined. Microglia, the brain's resident immune cells, are central to neuroinflammatory processes and may contribute to neuronal dysfunction in BPAN [2].

We report the first successful generation of microglia from BPAN patient-derived induced pluripotent stem cells (iPSCs), using established differentiation protocols and validation by canonical marker expression. Through a combination of targeted transcriptomics and secretomics, we uncovered that BPAN microglia display a unique, disease-associated phenotype—both at rest and after immune stimulation. Notably, secretomic profiling revealed unexpected shifts in the release of soluble factors, pointing to altered microglial communication and possible neurotoxic effects. Transcriptomic analysis further highlighted significant changes in pathways related to autophagy, apoptosis, microglial function, and both cytokine and inflammatory signaling. Together, these findings suggest that *WDR45* mutations may push microglia into a previously unrecognized pathogenic state, reshaping our understanding of BPAN's immune landscape. This patient-specific iPSC model opens new avenues for unraveling the complexities of neuroinflammation and for discovering therapeutic strategies to target microglial dysfunction in rare neurodegenerative diseases.

1 - Hayflick SJ, et al. β-Propeller protein-associated neurodegeneration: a new X-linked dominant disorder with brain iron accumulation. Brain. 2013 Jun;136(6):1708-17. doi: 10.1093/brain/awt095.

2 - Gao C, et al. Microglia in neurodegenerative diseases: mechanism and potential therapeutic targets. Signal Transduct Target Ther. 2023;8:359. doi: 10.1038/s41392-023-01588-0.

Abstract No. P116

Development of a hiPSC-based platform for high-throughput drug screening and disease modeling in hereditary pulmonary arterial hypertension

Clara Luise Plötner[1,2,*], *Sylvia Merkert*[1,2], *Till Janotte*[1], *Shannon A. McCallan*[1,2], *Laura Neuhaus*[1], *Tim Kohrn*[1], *Aileen Schmidt*[1,2], and *Ulrich Martin*[1,2]

[1] Leibniz Research Laboratories for Biotechnology and Artificial Organs (LEBAO), Department of Cardiothoracic, Transplantation and Vascular Surgery, MHH
[2] Biomedical Research in End-stage and Obstructive Lung Disease (BREATH), Member of the German Center for Lung Research (DZL), Hannover Medical School

*Presenting author

Pulmonary Arterial Hypertension (PAH) is a rare but severe cardiopulmonary disorder for which the underlying mechanisms remain incompletely understood. It is characterized by progressive vascular remodeling, leading to increased resistance, pulmonary arterial pressure, and ultimately right heart failure. Although Sotatercept has emerged as a promising new therapy targeting disease mechanisms, current treatments remain largely symptomatic and survival is still limited.

To identify additional therapeutic options, we are developing a high-throughput drug screening platform based on human induced pluripotent stem cells (hiPSCs) with a focus on hereditary PAH (hPAH). The most frequently mutated gene in hPAH is bone morphogenetic protein receptor type 2 (BMPR2), whose signaling through Smad1/5/8 normally induces anti-proliferative genes such as Inhibitor of Differentiation 1 (ID1). BMPR2 dysfunction disrupts signaling balance, decreasing Smad1/5/8 and increasing Smad2/3 activity, thereby promoting resistance to apoptosis, as well as proliferation and migration in vascular cells such as smooth muscle cells (SMCs).

To model these mechanisms *in vitro* and establish a scalable drug screening tool, we introduced an eGFP reporter into the ID1 locus of wild-type (WT) and patient-derived hiPSCs, generating ID1eGFP reporter lines. In addition, we created BMPR2 knockouts in both non-reporter WT hiPSCs and the WT-ID1eGFP line. All lines were differentiated into induced SMCs (iSMCs).

Reporter activity was assessed in pilot experiments using flow cytometry and plate reader measurements in a 96-well format. Genotype-dependent differences in ID1eGFP signal were observed, supporting the feasibility of the assay.

This platform integrates patient-derived, knockout and wild-type reporter lines to model hPAH pathomechanisms in iSMCs. We aim to determine whether these readouts provide a scalable basis for high-throughput drug screening to identify novel therapeutic candidates for hPAH.

Abstract No. P117

Single-cell profiling of striatal organoids derived from Leigh syndrome patients indicates an early imbalance of excitatory and inhibitory neurons during neurodevelopment

Martina Podlesnic[1,*], *Stephanie Le*[2], *Marta Suárez-Cubero*[1], *Antonio delSol*[3], *Alessandro Prigione*[2], and *Frank Edenhofer*[1]

[1] Institute of Molecular Biology, University of Innsbruck, Austria
[2] Department of General Pediatrics, Neonatology and Pediatric Cardiology, Heinrich Heine University Düsseldorf
[3] Luxembourg Centre for Systems Biomedicine (LCSB), University of Luxembourg

*Presenting author

Induced pluripotent stem cell (iPSC) derived organoids have emerged as powerful models to study the nervous system development *in vitro*. By recapitulating early neurodevelopment, brain organoids serve as valuable tools to study neurodevelopmental disease mechanisms. For this project we employ this tool to investigate the pathophysiology of Leigh syndrome (LS). LS is caused by mutations in mitochondrial genes leading to dysregulated ATP production. The nuclear DNA-encoded gene *NDUFS4* is frequently mutated in patients, causing alterations in various brain regions including the striatum. However, the underlying pathophysiological mechanisms driving this disease remain elusive. Here, we aim to investigate transcriptomic alterations during early striatal organoid development. Therefore, we employed iPSCs carrying a *NDUFS4* mutation and their isogenic controls. Single-cell transcriptomic profiling revealed an imbalance in neuronal subtype proportions of LS organoids. In particular, the fraction of glutamatergic neurons expanded at the expense of GABAergic neurons and cell type prioritisation highlighted GABAergic neurons as the most perturbed. Differential expression analysis confirmed downregulation of genes involved in OXPHOS on gene and pathway level with GABAergic neurons being highly susceptible to these changes. Across the vast majority of cell types, 23 shared dysregulated genes were found. Leveraging STRING analysis for network construction allowed us to further narrow down the DEG candidates to *NR2F1*, *FOXP2*, and *ETV1*. These genes are implicated in neural differentiation by PAX6 regulation, neurite outgrowth of striatal medium spiny neurons in mouse and regulation of GABAergic subunit in developing cerebellar neurons, respectively. Additionally, investigating the regulome using decoupleR, we found the majority of transcription factors regulating *NR2F1* and *ETV1* to be perturbed. In conclusion, our study revealed an early imbalance of excitatory and inhibitory neuronal subtypes and gene candidates involved in the distinct neuronal phenotypes in LS organoids, facilitating a more comprehensive understanding of LS pathology and establishing potential novel intervention paradigms.

Abstract No. P118

High throughput screening for compounds allowing readthrough of premature termination codons based on genetically engineered primary ciliary dyskinesia patient specific hiPSCs

*David Puertollano Martín**, *Laura von Schledorn*, *Theresa Niemöller*, *Nicole Cleve*, *Laurien Czichon*, *Sylvia Merkert*, *Felix Ringshausen*, *Ulrich Martin*, and *Ruth Olmer*

Hannover Medical School (MHH)

*Presenting author

Primary Ciliary Dyskinesia (PCD) is a rare genetic disease characterized by an impairment in the mucociliary clearance in the airways, which leads to recurrent respiratory infections, bronchiectasis and lung failure. It is caused by mutations in genes involved in cilia function, resulting in defective ciliated cells. Different mutations in more than 50 genes lead to PCD, generating a wide range of diseased phenotypes of which none can be currently cured. A notable number of all PCD-causing mutations are premature termination codons (PTCs), which can be potentially overcome by PTC suppressor therapies. Here, DNAI1-dTomato hiPSCs were generated via Cas9-based editing and further differentiated into proximal airway epithelia on air-liquid interface (ALI) cultures, where dTomato-fluorescent motile ciliated cells formed. A second cell line was generated by introducing a PCD patient-specific PTC in the DNAI1-dTomato construct in homozygosis. These cells, when differentiated on ALI cultures, formed immotile ciliated cells with a significantly reduced dTomato mean fluorescence intensity (MFI). This model was used to test known PTC suppressors on 96-well ALI inserts. After a 48-hour long treatment, Gentamycin, G418, and Escin, among others, significantly increased dTomato MFI. Differentiation into ciliated cells was also possible in submerged cultures on standard 96-well plates, paving the way for high throughput screening of compounds on 384-well plates. Of note, the feasibility to generate monoclonal reporter cell lines and the accurate representation of the PCD lung disease make this model a solid platform not only for drug screening, but also for testing other PTC suppresor therapies such as suppressor tRNAs.

von Schledorn, L, et al. (2023): Primary Ciliary Dyskinesia Patient-Specific hiPSC-Derived Airway Epithelium in Air-Liquid Interface Culture Recapitulates Disease Specific Phenotypes In Vitro. Cells 12(11)

Toledano, I (2024). Genome-scale quantification and prediction of pathogenic stop codon readthrough by small molecules. Nature Genetics 56

Abstract No. P119

Genetic screen to identify lipid metabolism regulators using hiPSC-derived hepatocytes

Merrit Romeike[1,2,*], *Ferhat Ali Yaman*[1,2], *Fabian Bachinger*[1,2], *Dominika Dziedzicka*[3], and *Ludovic Vallier*[1,2]

[1] Berlin Institute of Health (BIH), Berlin
[2] Max Planck Institute for Molecular Genetics, Berlin
[3] Wellcome-MRC Cambridge Stem Cell Institute, Cambridge, UK

*Presenting author

Metabolic dysfunction-associated steatotic liver disease (MASLD) is one of the most common liver diseases worldwide. MASLD is a progressive disease, which goes through stages including cell death, inflammation, fibrosis, cirrhosis and hepatocellular carcinoma or liver failure. Liver transplantation remains the only available therapy, but due to the lack of organ donors only a limited number of patients can benefit from this approach. The first step of MASLD progression is characterized by cell death of hepatocytes, the main parenchymal cell type of the liver. There are yet no pharmacological strategies available to protect hepatocytes from lipid toxicity and other cellular stress. Thus, there is an urgent need to identify novel druggable targets to protect hepatocytes against excessive accumulation of lipids.

To address this challenge, we are developing a pooled scalable CRISPR-KO screening strategy to identify novel regulators of lipid metabolism in liver cells. As *in vitro* culture of human hepatocytes remains challenging, we took advantage of our protocol for generating hepatocytes by using forward programming of hiPSCs into FoP-hepatocytes. We demonstrate the suitability of FoP-hepatocytes as a disease model for MASLD by inducing steatosis through free fatty acid treatment and recapitulating the role of known regulators of lipid metabolism and genetic risk factors for MALSD. We are now combining disease modeling with targeted sub-genomic and genome-wide lentiviral CRISPR-KOs. Staining of intracellular neutral lipids allows flow cytometry-based sorting of hepatocytes with increased and decreased steatosis. Subsequent analysis will identify positive and negative regulators of hepatic lipid metabolism.

Together, our results describe a new hiPSC-derived platform to perform genetic screens and to identify mechanisms driving hepatocyte injury in the context of liver diseases.

Abstract No. P120

Psilocin fosters neuroplasticity in iPSC-derived human cortical neurons

Malin Schmidt[1,2,*], *Anne Hoffrichter*[1,2], *Mahnaz Davoudi*[1,2], *Sandra Horschitz*[1,2], *Thorsten Lau*[1,3], *Marcus W. Meinhardt*[1], *Rainer Spanagel*[1], *Julia Ladewig*[1,2], *Georg Köhr*[3], and *Philipp Koch*[1,2]

[1] Central Institute of Mental Health (ZI), Mannheim
[2] German Cancer Research Center (DKFZ)
[3] Mannheim Centre for Translational Neuroscience (MCTN)

*Presenting author

Psilocybin is under investigation as a novel treatment for anxiety, substance use disorders, and treatment-resistant depression. Preclinical studies suggest that psychedelics enhance neuronal plasticity by strengthening synaptic responses and promoting protein synthesis. However, the molecular and cellular mechanisms underlying psilocybin's effects in the human brain remain largely unknown.

Here, we exposed human cortical neurons derived from induced pluripotent stem cells to psilocin, the active metabolite of psilocybin and a 5-HT2A receptor agonist. We examined gene expression, neuronal morphology, synaptic protein abundance, and functional activity. Psilocin treatment led to 5-HT2A receptor–mediated upregulation of BDNF, accompanied by transcriptomic signatures indicative of enhanced neuroplasticity. Morphological analyses revealed increased neuronal complexity and elevated expression of synaptic proteins, particularly within postsynaptic compartments. Functionally, psilocin exposure increased neuronal excitability and strengthened synaptic network activity.

Together, these findings suggest that psilocin induces a state of heightened neuronal plasticity in human neurons, providing a mechanistic basis for its therapeutic potential in neuropsychiatric disorders associated with synaptic dysfunction.

Abstract No. P121

Network hyperexcitability and dopaminergic neuron loss in a novel A30P-driven Parkinson's midbrain organoid model

Kea Aline Schmoll[*], *Ole Jensen*, *Emre Taylan Duman*, *Guobin Bao*, *Pratulya Shah*, *Eunseo Kim*, *Michael Gani Setya*, *Branimir Berecic*, *Wolfram-Hubertus Zimmermann*, and *Maria-Patapia Zafeiriou*

University-Medical Center Goettingen

*Presenting author

Midbrain dopaminergic (mDA) neurons regulate motor control, reward, and emotion, and are selectively vulnerable in Parkinson's disease (PD). Current therapies provide transient symptomatic relief but fail to halt disease progression, as treatment efficacy declines with continued neuronal loss. Despite intense research, the mechanisms underlying mDA degeneration remain poorly understood, underscoring the need for novel therapeutic strategies. To address this, we developed midbrain -enriched neuronal organoids (mBENOs) from human induced pluripotent stem cells. mBENOs display a reproducible architecture comprising bona fide mDA neurons embedded within a functional neuronal-glial network that includes astrocytes, oligodendrocytes, and microglia. Whole-mount immunofluorescence and temporal transcriptomic analysis confirmed the co-development of dopaminergic, GABAergic, glutamatergic, and cholinergic neurons. Temporal analysis of local field potential recordings demonstrated increasing spontaneous activity, supporting functional maturation. To model PD, we introduced the SNCA A30P mutation via genome engineering. SNCA A30P mBENOs retained differentiation capacity but exhibited progressive mDA neuron loss from day 60 onward and hyperexcitability resembling basal ganglia dysfunction. Together, these findings establish the mBENO as a robust and scalable human model for dissecting PD pathomechanisms, tracking disease progression, and enabling preclinical therapeutic screening.

Abstract No. P122

Mitochondrial DNA editing enables the development of iPSC models of maternally inherited Leigh syndrome

Isabella Tolle[1,*], *Claudio Fiorini*[2], *Jochen Dobner*[3], *Pawel Lisowski*[4,5], *Emanuela Bottani*[6], *Valerio Carelli*[2], *Andrea Rossi*[3], and *Alessandro Prigione*[1]

[1] Medical Faculty, Heinrich Heine University Düsseldorf
[2] University of Bologna, Italy
[3] IUF-Leibniz Research Institute for Environmental Medicine, Düsseldorf
[4] Max Delbrück Center for Molecular Medicine in the Helmholtz Association (MDC), Berlin
[5] Institute of Genetics and Animal Biotechnology, Polish Academy of Sciences, Warsaw, Poland
[6] University of Verona, Italy

*Presenting author

Engineering mitochondrial DNA (mtDNA) has been historically challenging. Inefficient transport of nucleic acids into mitochondria and lack of efficient double-strand break repair prevent the use of CRISPR/Cas in mitochondria. The multi-copy nature of mtDNA further complicates the study of mitochondrial disorders, resulting in a paucity of effective model systems and cures. One of the most severe mtDNA disorders in children is MILS (Maternally inherited Leigh syndrome), which affects the central nervous system causing psychomotor regression and early death. Here, we applied a recently developed mtDNA editing tool termed DddA-derived cytosine base editor (DdCBE) to human induced pluripotent stem cells (iPSCs) in order to generate innovative MILS models. We focused on two MILS-causing variants in the genes *MT-ND1* and *MT-ND5*, both belonging to Complex I (CI) of the mitochondrial respiratory chain. After having selected the most effective constructs, we transfected them into the healthy control iPSC line BIHi005-A-24, harboring a NGN2 cassette for rapid neuronal generation. We were able to obtain clonal iPSC lines of BIHi005-A-24 with up to 82% on-target editing, minimal off-target editing and stable mutation levels during passaging. For each target site we chose three lines with varying degrees of editing (16%-73% for *MT-ND1*, 51%-82% for *MT-ND5*) for further experiments. We are currently assessing CI activity and the mitochondrial membrane potential in the edited iPSC lines to determine the extent of impairment caused by the introduced variants. We differentiated the edited lines into NGN2 neurons and are using bulk transcriptomics to build mutation-related signatures and are carrying out high-content analysis to quantify neuronal branching capacity. To our knowledge, this is the first report demonstrating the generation of MILS neuronal models using mtDNA editing. These new models could play an important role in shedding light on the pathogenetic mechanisms of MILS and possibly pave the way for therapeutic developments.

Abstract No. P123

Human iPSC-derived BBB model for CNS-targeted PROTAC discovery

Arjen Weller[1,*], *Daniel Stach*[1], *Undine Haferkamp*[1], *Johanna Huchting*[1], *Patricia Muschong*[2], *Mario Mezler*[2], and *Ole Pless*[1]

[1] Fraunhofer ITMP, Frankfurt a. Main, Discovery Research ScreeningPort Hamburg
[2] AbbVie Deutschland GmbH & Co. KG, Wiesbaden

*Presenting author

Neurodegenerative diseases are characterized by the toxic accumulation of misfolded and aggregated proteins, which are traditionally considered hard to drug with conventional small-molecule inhibitors. Proteolysis-targeting chimeras (PROTACs), a class of molecular degraders, offer a promising therapeutic strategy by leveraging the ubiquitin–proteasome system to induce the selective degradation of target proteins, rather than merely inhibiting their function. This approach enables the targeting of previously undruggable proteins that evade classical drug modalities. However, the development of central nervous system (CNS) therapies using PROTACs is significantly hindered by their high molecular weight and complex physicochemical properties, which limit their ability to cross the blood–brain barrier (BBB).

To address this challenge, we developed an advanced in vitro BBB model based on human-induced pluripotent stem cell-derived brain capillary endothelial cells (hiPSC-derived BCECs). Unlike conventional models, these BCECs exhibit in vivo-like BBB characteristics, including the expression of a broad spectrum of marker proteins, restricted transcellular transport, and high transendothelial electrical resistance. Additionally, a receiver cell line expressing the target protein was integrated into the BBB model to enable direct quantification of transported PROTACs and their degradation efficiency. In this study, we focused on leucine-rich repeat kinase 2 (LRRK2) as a target, as mutations in LRRK2 are associated with an increased risk of Parkinson's disease, making it a critical therapeutic target. Here, we present our physiologically relevant BBB co-culture model for evaluating and optimizing the BBB permeability and degradation efficiency of the LRRK2 degrader XL01126.

Abstract No. P124

Modeling and reversing Alzheimer's-associated microglial states using iPSC-derived cells and disease-linked gene signatures

Henrik Renner, *Tim Yang*, *Fabrizia Zoppetti*, *Alfonso Gerado Garcia Diaz Barriga*, *Georgina Penalosa-Ruiz*, *Benjamin Fischer*, *Ane Iturbide Martinez de Albeniz*, *Wynona Sophie Mülle*[1], *Stefanie Maedje*, *Isabelle Meyer*, *Tayfun Kilic*, *Daniel Rosebrock*, *Judith Thoma*, *Carina Wollnik*, *Anastasia Illarionova*, *Philip Peschutter*, *Hanna Schubert*, *Alexandra Hackmann*, *Mark Gemkow*, *Rainer Kuhn*, *Sandra Lubitz*, *Bastian Zimmer*[*], and *Doris Gruber-Schoffnegger*

Evotec SE, Hamburg

*Presenting author

Microglia are critical for maintaining brain homeostasis and orchestrating immune responses in the central nervous system. Dysregulation of microglial function is increasingly implicated in the pathogenesis of Alzheimer's disease (AD), yet the molecular mechanisms underlying this dysfunction remain incompletely understood. While transient microglial activation may confer neuroprotection, chronic exposure to pathological stimuli—such as amyloid-β (Aβ) plaques and hyperphosphorylated tau—drives microglia toward a dysfunctional, unresponsive state.

In this study, we identified an AD-associated microglial gene expression signature by analyzing post-mortem brain tissue from over 650 individuals, including AD patients and age-matched controls. This signature reflects a transition from homeostatic to dysregulated microglial states and correlates with disease burden and progression.

To model this phenotype in vitro, we developed a human induced pluripotent stem cell (iPSC)-derived microglia system that recapitulates the transcriptional profile of AD-associated microglia. Functional assays revealed hallmark impairments, including diminished phagocytic capacity and disrupted lipid metabolism.

Using this platform, we performed a small molecule screen to identify compounds capable of restoring microglial homeostasis using transcriptomics as a readout. Several candidate modulators emerged, one of which successfully reversed key transcriptional alterations and rescued microglial function.

Our finding suggests that (iPSC)-derived microglia systems that recapitulate AD-associated microglial states may have potential for therapeutic discovery in Alzheimer's disease.

Collaborative work with Bristol Myers Squibb (BMS)

P0125 – P144: Stem cells in regenerative therapies

P125 Mef2c promotes the maturation of Ascl1-induced basket cell-like GABAergic neurons
Filippo Calzolari

P126 Feeder-free equine embryonic fibroblast reprograming to iPSCs
Rezwan Firuzi

P127 Investigation of signaling cues driving directed RPE specification from human iPS cells
Boris Greber

P128 Enhancing adherent stem cell culture with the CellScrew®: A scalable and sustainable system for bioprocessing
Katharina Günther

P129 New off-the-shelf GMP-compliant iPS cells and banking process for application in cell therapy
Björn Hiller

P130 Development of GMP-compatible genetically modified iPSC-derived macrophages for off-the-shelf cell therapy
Alica Hinkelmann

P131 The age of hiPSC-derived cardiomyocytes influences their contractile synchronisation in a 3D engineered heart tissue model
Vojtech Hradil

P132 A new CGMP compliant adherent PSC culture medium that enables robust performance in cell therapy development
Katja Hufschmid

P133 Generating immune stress-resistant β cells for cell replacement therapy
Sophie Kipper

P134 Novel strategies to enhance magnetic cell targeting
Lisa Münchhalfen

P135 Mass production of lung epithelial progenitor cells from human iPSCs for regenerative approaches
Theresa Niemöller

P136 Utilizing CIMmultus OH™ for efficient purification of MSC EVs
Valentina Novak

P137 Exploring cardiac repair mechanisms with isolated and immortalized epicardium-derived cells
Katja Nowak

P138 Optimizing pluripotent stem cell spheroid growth in liter scale bioreactors through the use of constant medium perfusion
Sara Perez Munoz

P139 Off the shelf immune-shielded stem cell-derived γδ T cells for clinical applications
Tommaso Pezzica

P140 Clinical scale non-viral gene editing and feeder-free production of CAR-NK cells
Corinna Pohl

P141 Clonal and lineage tracing of barrier mesothelium
Aydan Sardogan

P142 Retrospective gene expression analysis in neural stem cells
Zeynep Aydin

P143 ELAPOR2 regulates proliferation of pancreatic progenitors and endocrine differentiation
Osvaldo Rodrigues Pereira

P144 Extraction by AI for tracking of hPSC-based clinical trials
Meiying Xue

Abstract No. P125

Mef2c promotes the maturation of Ascl1-induced basket cell-like GABAergic neurons

Filippo Calzolari[1,*], and *Benedikt Berninger*[1,2]

[1] Institute of Physiological Chemistry, University Medical Center JGU Mainz
[2] Centre for Developmental Neurobiology and MRC Centre for Neurodevelopmental Disorders Institute of Psychiatry and Neuroscience, King´s College London, UK

*Presenting author

Fate conversion of non-neuronal cells by overexpression of neurogenic transcription factors (TFs) holds promise as a source of induced neurons for basic and translational research purposes. While neural progenitor-specific TFs such as Ascl1 can efficiently reprogram neural and non-neural cells to induced neurons (iNs), our ability to instruct the acquisition of molecular, morphological and functional properties typical of endogenous neurons is still limited. Overexpression of a phosphorylation-defective mutant form of Ascl1 (Ascl1SA6) has recently been shown to convert cortical glia to an inhibitory neuron-like identity with marker expression (e.g. Parvalbumin) and functional features typical of cortical fast-spiking interneurons such as Basket cells, while retaining broadly immature morphological characteristics. While encouraging, these observations suggest that expression of TFs with endogenous roles at later stages of neuronal differentiation may be necessary to promote iN maturation. To test this hypothesis, we overexpressed Mef2c, a TF essential for Basket cell differentiation and whose expression is maintained in mature neurons. Intriguingly, while overexpression of Mef2c alongside Ascl1SA6 did not affect the abundance of Parvalbumin-expressing iNs, these neurons displayed more complex morphologies and progressively acquired the ability to extensively decorate the soma and dendrites of nearby iNs, resembling endogenous Basket cells. Interestingly, such morphologies were associated with limited expression of markers of mature GABAergic synapses in vitro, suggesting that complex morphological and cell-cell interaction programs can be activated independently of synaptogenesis in the course of reprogramming. These observations highlight the modular nature of neuronal differentiation following fate conversion. Finally, in vitro analyses of reprogramming factor expression levels and in vivo reprogramming outcomes indicate that while elevated Ascl1SA6 expression levels may be necessary to efficiently drive reprogramming, they may hinder certain aspects of maturation, which may require novel approaches to reprogramming factor expression control.

Reprogramming astroglia into neurons with hallmarks of fast-spiking parvalbumin-positive interneurons by phospho-site–deficient Ascl1; 10.1126/sciadv.adl5935

Abstract No. P126

Feeder-free equine embryonic fibroblast reprograming to iPSCs

Rezwan Firuzi[*], *Kurt Pfannkuche,* and *Sarkawt Hamad*

Marga-and-Walter-Boll-Laboratory for Cardiac Tissue Engineering, University of Cologne

*Presenting author

The potential of human and animal-induced pluripotent stem cells (iPSCs) extends to the domains of regenerative medicine and disease modelling, including veterinary applications. However, the generation of induced pluripotent stem cells (iPSCs) has become a standard protocol in most laboratories and is currently employed in clinical trials. In contrast, the generation of equine iPSCs (EiPSCs) has been hindered by the limited efficiency and suboptimal culture conditions. In this study, we sought to optimize the conditions for feeder-free culture of EiPSCs. We reprogrammed equine fibroblast cells to iPSCs using episomes and transposons. The culture was observed for a 36-day period following induction, and flow cytometry analysis was implemented to ascertain the efficiency of reprogramming in comparison to non-reprogrammed equine fibroblast.

Cell morphology was altered during reprogramming. Furthermore, flow cytometry revealed expression of OCT3/4 in a large fraction of cells. The utilization of our workflow supports feeder- and serum-free culture of equine embryonic fibroblast and their reprogramming into EiPSCs under defined conditions.

Keyword: Equine embryonic fibroblast, Feeder-free, Equine reprograming

Abstract No. P127

Investigation of signaling cues driving directed RPE specification from human iPS cells

Elitsa Borisova[1,2], *Soraia Martins*[1], *Thomas Berger*[1], *Daniel Terheyden-Keighley*[1], and *Boris Greber*[1,*]

[1] Catalent Düsseldorf
[2] Julius-Maximilians-Universität Würzburg

*Presenting author

The retinal pigment epithelium (RPE) is essential for maintaining retinal health. Its dysfunction causes degenerative disorders leading to vision impairment. Currently, cell replacement is the only approach offering regeneration and functional improvement in many of these diseases. iPSC-derived RPE offers the potential for scalable and off-the-shelf allogeneic therapy. Production of such cells requires robust quality control, including assays testing raw material functionality. This is challenging when their mechanism of action is unclear, as with Activin A - a signaling factor widely used for RPE induction.

A raw material activity assay for a fully GMP-compliant RPE differentiation protocol was developed. The method relies on transient inhibition of FGF, TGF-β, and BMP signaling with small molecules, combined with long-term Activin A supplementation. Factor withdrawal experiments analyzed with RT-qPCR identified early marker genes reporting on the activity of each factor. To further clarify the role of Activin A in driving RPE fate, time course-based RNA sequencing in Activin-treated versus Activin withdrawal conditions was performed.

SPRY4, LEFTB, and HAND1 were identified as reliable markers for the FGF, TGF-β, and BMP pathway inhibitors, respectively. LEFTB also responded to Activin A but only when small molecules were omitted to avoid interference. Subtractive analysis of the RNA-sequencing time course revealed consistent upregulation of markers reflecting activation of SMAD2/3 signaling, as well as RPE differentiation upon Activin A treatment. Moreover, the data could be used to uncover the earliest events of Activin A stimulation in RPE specification to generate hypotheses about the mode of action.

The developed assay enables efficient testing of four key raw materials in RPE production by directly assessing their activity on iPSCs. Analysis of the role of Activin A revealed new potential markers and mechanisms underlying RPE fate induction, improving quality control in GMP-grade RPE manufacturing and offering new insights into human developmental biology.

Abstract No. P128

Enhancing adherent stem cell culture with the CellScrew®: A scalable and sustainable system for bioprocessing

Björn Boshof, *Annika Herchenröder*, *Paul Medebach*, *Katharina Günther*[*], *Joel Eichmann*, and *Lukas Käßer*

Green Elephant Biotech GmbH, Gießen

*Presenting author

The increasing demand for large-surface-area adherent cell culture platforms in stem cell-based applications calls for innovative and scalable solutions. Human induced pluripotent stem cells (hiPSCs) and their differentiated lineages require robust, scalable, and standardized culture systems that enable high-quality cell production while minimizing labor intensity and variability. The CellScrew® is a novel, single-use cultivation system with a surface from 850cm ^{2} to 10,000 cm^2, 3D-printed from plant-based polylactic acid (PLA). Its patented design of concentric cylinders and Archimedes' screws provides a high surface-to-volume ratio, enabling efficient large-scale cell culture. This architecture supports efficient nutrient and oxygen distribution, enabling reproducible adherent cell growth while reducing the environmental carbon footprint by up to 90% compared to conventional polystyrene-based culture ware.

Initial evaluation of the CellScrew® 6K using HEK293 cells confirmed robust cell growth with high cell recovery of more than 90% achievable at reduced trypsin concentrations, underscoring the system's gentle harvesting properties. Based on these findings, the platform is now being applied to hiPSC workflows. The CellScrew® supports expansion of high quality hiPSCs for master and working cell banking, large-scale production of iPSC-derived cells, and subsequent differentiation into clinically relevant lineages including cardiomyocytes and other lineages. These features position the technology as a valuable tool for both basic research and translational applications in cell therapy, regenerative medicine and tissue engineering. The availability of both RUO and GMP-compatible versions ensures continuity across the translational pipeline, from laboratory research through to therapeutic manufacturing.

By combining scalability, handling efficiency, and sustainability, the system addresses key bottlenecks in stem cell manufacturing. Looking ahead, integration with an automation platform is under development, further reducing manual workload and enabling standardized large-scale stem cell bioprocessing.

Abstract No. P129

New off-the-shelf GMP-compliant iPS cells and banking process for application in cell therapy

*Björn Hiller**, *Claudia Träger*, *Noah Henschel*, *Jens Lichte*, *Anna Gamerschlag*, *Melanie Huehne*, *Francois Hafezi*, *Agnes Beermann*, *Andrea Meffert*, and *Boris Greber*

Catalent Düsseldorf GmbH

*Presenting author

Cell therapy has evolved as an important instrument for the treatment of many life-threatening diseases. Induced pluripotent stem cells (iPSCs) have been shown to be particularly valuable as starting material for the downstream differentiation of various cell types and their application in cell therapy. However, the generation of GMP-compliant, clinical-grade iPSCs is challenging, as expansion protocols need to be optimized to prevent suboptimal culture conditions, to reduce overall cell passagings and to avoid the risk of acquiring increased mutation loads. At the same time, expansion needs to allow the cryopreservation of large cell banks requested by the customers. To this end, we have developed a GMP-compliant process for the banking of induced pluripotent stem cells (iPSCs). For iPSC expansion and cryopreservation, we have recently established and validated a new process based on single cell passaging that allows fully controllable expansion and subsequent cryopreservation of up to 360 vials with 2x10^6 cells each. Based on this process, we have generated several EMA/FDA-compliant iPSC master cell banks (MCBs) with full informed consent of German or American origin that are available off-the-shelf.

Furthermore, we have established a comprehensive GMP-compliant quality control (QC) framework to ensure the safety, identity, purity, and potency and to meet current EMA/FDA requirements. Safety testing includes microbial and viral assessments to confirm the absence of contaminants. Identity and purity are verified by iPSC-specific marker expressions. Genetic stability is monitored to detect chromosomal abnormalities or other genomic alterations. Potency is assessed by confirming the cells' ability to differentiate into relevant lineages and maintain functional integrity. This integrated QC approach ensures consistency, regulatory compliance, and suitability of the iPSC MCB for downstream applications.

MCB-derived research cell banks (RCBs) have also been generated for each MCB, are readily available and highly suitable for testing in the individual application by each potential partner.

Abstract No. P130

Development of GMP-compatible genetically modified iPSC-derived macrophages for off-the-shelf cell therapy

*Alica Hinkelmann**, *Philine Scheinpflug**, *Kathrin Haake*, *Andrej Schebet*, *Elisabeth Derksen*, *Farah Aqel*, *Jonas Klaas*, *Tim Magiera*, *Sebastian Kirsch*, *Anais Duval*, *Philip Hublitz*, *Michela Mirenda*, *Martin Briscadieu*, *Michael Esquerre*, *Michael Epstein*, *Katrin Ridders*, *Ariane Walsh*, *Stefanie Pfaender*, *Andreas Scheel*, *Matthias Austen*, and *Audrey Holtzinger*

Evotec SE, Göttingen

*Presenting authors

Recently, autologous macrophages have gained increasing attention for treatment of solid tumors and fibrotic diseases due to their potential to deeply infiltrate the tissue, adapt to their environment and key role in coordinating immune responses. Their therapeutic potential as cell therapeutics can be enhanced by genetic engineering—for example, by introducing chimeric antigen receptors (CARs) for antigen-specific targeting, deleting checkpoint inhibitors to boost activity, or modifying HLA molecules to evade immune detection. Production of autologous macrophages for clinical and commercial application remains challenging due to limited patient material, intricate genetic manipulations, and manufacturing complexity. Contrary to this, iPSC-derived macrophages (iMACs) from clonal genetically engineered iPSC show advantages regarding availability, modification possibilities and stringent quality control.

GMP-compatible iPSCs were edited with a targeted approach to generate iMACs with modified functionality: lacking the checkpoint inhibitor SIRPα to increase phagocytosis, expressing a CAR to target fibrotic or tumor cells and immune shielding to increase persistence. Following genetic engineering, iPSCs were enriched and clonally expanded. Resulting cell lines underwent quality control to verify transgene integration, genomic stability, and preservation of pluripotency. Cells were differentiated using a scalable 3D xeno-free process allowing large-scale production. Fully functional macrophages with a purity of >90% were obtained and transition to GMP-compatible reagents increased fold expansion significantly.

SIRPα knockout resulted in increased phagocytosis and cytotoxicity in differentiated macrophages. Likewise, CAR expression and functionality were validated, demonstrating enhanced uptake and killing of antigen-expressing target cells. Feasibility of generating immune-shielded iMACs has been demonstrated.

In summary, we have developed clonal iPSC lines incorporating CARs, SIRPα deletion, and immune evasion edits to generate highly pure, off-the-shelf GMP-compliant iMACs tailored for the treatment of solid tumors and lung or liver fibrosis.

Abstract No. P131

The age of hiPSC-derived cardiomyocytes influences their contractile synchronisation in a 3D engineered heart tissue model

Vojtech Hradil

Leibniz Research Laboratories for Biotechnology and Artificial Organs (LEBAO), Hannover

Human induced pluripotent stem cells (hiPSCs) can be differentiated into functional cardiomyocytes (CMs) for *in vitro* assays or *in vivo* application. For both, it is crucial that their physiological properties are maintained after differentiation. Recent pre-clinical studies revealed that the application of aggregated hiPSC-derived CMs to the infarcted myocardium leads to functional recovery, yet with occurring arrhythmias as a side-effect, possibly due to insufficient engraftment and imperfect coupling. Therefore, we decided to study the capacity of aggregated hiPSC-derived CMs to functionally synchronise with each other *in vitro* in a 3D engineered heart tissue model.

A transgenic hiPSC line with the genetically encoded calcium indicator GCaMP6f was differentiated into ventricular-like cardiomyocytes in the form of aggregates (hiCMAs). hiCMAs, together with human foreskin fibroblasts and rat collagen-based matrix components, were used to produce bioartificial cardiac tissues (BCTs), a well-established scaffold-based engineered heart tissue model for studying cardiac (patho)physiology *in vitro*. BCTs were cultivated for 21 days with a progressive stretch from day 7 onwards to support tissue maturation. Tissues with similar hiCMA density were periodically monitored via fluorescence microscopy to track calcium fluctuations. Obtained videos were analysed using a novel, semi-automated approach to follow individual hiCMAs over time and analyse the progress of their synchronisation.

To achieve synchronisation of 50% of hiCMAs, typically 3-4 days were required, regardless of hiCMAs‘ age (ranging from 4 to 9 weeks). However, to reach the point of full synchronisation of all the aggregates, older hiCMAs (> 7 weeks) required approximately 1.5-2 fold more time to fully synchronise with each other compared to their younger counterparts. Moreover, a higher variance in the time point of full-tissue synchronisation has been observed when older hiCMAs were used. To further elaborate on the synchronisation differences, we will investigate the underlying changes in cell-cell contacts via gene expression analysis of BCTs.

Abstract No. P132

A new CGMP compliant adherent PSC culture medium that enables robust performance in cell therapy development

Marcus Bunn[1], *Lauren Sangenario*[1], *Rebecca Guice*[1], *Michael Akenhead*[1], *Navjot Kaur*[1], *Mark Kennedy*[1], *Katja Hufschmid*[2,*], and *David Kuninger*[1]

[1] Thermo Fisher Scientific, Frederick, US
[2] Thermo Fisher Scientific, Reinach, Switzerland

*Presenting author

Culture systems for human pluripotent stem cell (PSC) expansion enable generation of a nearly unlimited pool of cells capable of differentiation to all three germ lineages with potential for cell-based regenerative therapies. To advance PSC therapy research to the clinic, there is a need for high-quality ancillary materials with GMP-compliant manufacturing, quality and safety testing, raw material traceability and supporting regulatory documentation. To ease this transition, we have developed Gibco Cell Therapy Systems (CTS) StemFlex Medium based on research use StemFlex Medium, but without raw materials directly of human or animal origin. CTS StemFlex Medium supports expansion of high-quality PSCs as well as challenging applications in the PSC workflow. Here we demonstrate PSCs cultured long-term in CTS StemFlex Medium maintain typical PSC characteristics including >95% expression of OCT4/NANOG pluripotency markers, normal morphology, trilineage differentiation potential and karyotype. This new medium allows for versatility in culture conditions: a flexible feed schedule with a weekend-free option and compatibility with multiple defined matrices and passaging reagents. CTS StemFlex Medium is shown to provide critical support of stressful PSC applications including single-cell passaging, with consistent cumulative fold expansion over 10 passages, maintenance of normal karyotype and >95% OCT4/NANOG expression. This new medium supports PSC expansion post electroporation-based delivery of the CRISPR Cas9-gRNA complex as well as downstream clonal expansion of edited PSCs. Adherent PSCs cultured in CTS StemFlex Medium can be transitioned to 3D culture using CTS StemScale PSC Suspension Medium resulting in high-quality PSC spheroids.

Abstract No. P133

Generating immune stress-resistant β cells for cell replacement therapy

Sophie Kipper[1,*], *Ingo Burtscher*[1], *Corinna Cozzitorto*[1], *Lorenzo Pasquali*[2], and *Heiko Lickert*[1]

[1] Helmholtz Munich
[2] Universitat Pompeu Fabra (UPF), Barcelona, Spain

*Presenting author

Type 1 diabetes (T1D) is an autoimmune disorder resulting in the destruction of insulin-producing β cells. $CD4^+$ T cells play a crucial role in the manifestation of autoimmunity as well as in graft rejection, in the case of transplanted islets. Stem cell-derived β (SC β) cells have paved the way for restoration of glycaemic control in patients after transplantation of *in vitro* generated stem cell-derived islets (SC-islets), circumventing the shortage of cadaveric islets. However, the immune system still poses a threat to transplanted SC-islets. This study aims to generate SC-islets resistant to immune-related stress.

Our collaborators in the Pasquali laboratory established a co-culture system using EndoC-βH1 cells or human islets and activated $CD4^+$ T cells. Epigenetic changes identified in β cells upon co-culture were aligned to single nucleotide polymorphisms (SNPs) associated with T1D from the Genome Wide Association Studies (GWAS). Activating Transcription Factor 3 (ATF3), whose role remains ambiguous in the context of β cells, was identified as a possible target gene involved in immune-related stress response. We created an ATF3 knock out iPSC cell line to test this hypothesis.

In parallel, we adapted the co-culture to our SC-islets system to corroborate the findings from EndoC-βH1 cells and human islets. Mimicking the presence of $CD4^+$ T cells, we tested concentrations of cytokines released by activated $CD4^+$ T cells and assessed their downstream effects. We will repeat these experiments with the ATF3 knock out cells and activated T1D-derived $CD4^+$ T cells. Our results indicate that manipulation of ATF3 could affect β cell response to immune-related stress. By generating SC β cells resistant to immune-related stress, we could support the advancement in the field of β cell replacement therapy for T1D.

Abstract No. P134

Novel strategies to enhance magnetic cell targeting

Lisa Münchhalfen[1,*], *Sarkawt Hamad*[2], and *Kurt Pfannkuche*[2]

[1] Marga and Walter Boll Laboratory for Cardiac Tissue Engineering, Cologne
[2] Institute for Neurophysiology, University of Cologne

*Presenting author

The potential of induced pluripotent stem cells has created many applications in the field of cell therapy. Some applications, especially those in the field of cardiac replacement therapies, suffer from low efficiencies of cell engraftment. Therefore, magnetic cell targeting can be discussed as a future perspective for capturing cells in the tissue.

Two methods of magnetic cell targeting are employed: the intracellular loading with superparamagnetic iron oxide nanoparticles (SPIONs), as well as cell labelling with ferromagnetic particles. SPIONs loading of cells using magnetic force mediated endocytosis, was already well described by Zhang (2023). We propose novel strategy of bio-linking ferromagnetic particles to the cell surface using antibodies.

Both methods for magnetically targeting cells are compared for their adhesion kinetics and clustering potential inside the magnetic field.

The adhesion assays demonstrate a faster adhesion kinetic of SPIONs-loaded MSC spheroids when a magnetic field was applied. SPIONs-loaded and ferromagnetic-particle-loaded cells performed equally in the cell clustering assay.

[1] Zhang, L. et al. Force-Mediated Endocytosis of Iron Oxide Nanoparticles for Magnetic Targeting of Stem Cells. Appl. Mater. Interfaces 2023, 15, 50574–50585.

Abstract No. P135

Mass production of lung epithelial progenitor cells from human iPSCs for regenerative approaches

Theresa Niemöller[1,2,*], *Sylvia Merkert*[1,2], *Ulrich Martin*[1,2], and *Ruth Olmer*[1, 2]

[1] Leibniz Research Laboratories for Biotechnology and Artificial Organs (LEBAO), Hannover Medical School
[2] Biomedical Research in End-stage and Obstructive Lung Disease (BREATH), Member of the German Center for Lung Research (DZL), Hannover Medical School

*Presenting author

Chronic lung diseases such as idiopathic pulmonary fibrosis and chronic obstructive pulmonary disease lack curative treatments, which highlights the urgent need for innovative therapeutic options. Human induced pluripotent stem cells (hiPSCs) are a promising source for generating lung epithelial cells for cell therapies and drug screening applications. To date, 2D conditions limit mass production of differentiated epithelial cells. This project aims to establish scalable, 3D culture conditions to generate hiPSC-derived lung epithelial progenitor cells of high quality and quantity.

Differentiation of lung epithelial cells in adherent culture is achieved by following a well-established stepwise differentiation protocol from definitive endodermal (DE) cells via anterior foregut endoderm (AFE) to early lung progenitors (LP), and ultimately to proximal/distal lung epithelial cells. DE cell generation from hiPSCs has already been successfully adapted to 3D scalable culture conditions, including stirred tank bioreactors. Subsequent differentiation steps now need to be transferred to scalable formats by investigating key culture factors such as cell density, format, reaggregation, timing, and concentration of signaling molecules. Flow cytometry is applied to analyze marker expression of DE, AFE, and LP cells. A CRISPR/Cas9-engineered NKX2.1-eGFP reporter line enables real-time monitoring of LP differentiation.

We demonstrate a successful differentiation of hiPSCs towards lung progenitor cells in suspension culture, which is confirmed by the expression of CXCR4, cKit, EpCAM (DE), FOXA2 (AFE) and NKX2.1 (LP). Importantly, we observed that adjusting reaggregation conditions and cell densities substantially increases the proportion of NKX2.1$^+$ lung progenitors. Current efforts focus on further enhancing the yield of NKX2.1$^+$ progenitors by refining differentiation conditions. These findings demonstrate a proof of principle for scalable differentiation of hiPSCs into lung epithelial progenitor cells. Establishing a robust, scalable protocol for hiPSC-derived lung differentiation will provide a foundation for future cell-based therapies and high-throughput drug screening platforms targeting chronic lung diseases.

Abstract No. P136

Utilizing CIMmultus OH™ for efficient purification of MSC EVs

Valentina Novak, *Andrej Raspor*, *Darja Bozic*, *Ivana Petrovic Koshmak*, *Katja Vrabec*, *Maja Leskovec*, and *Ales Strancar*

Sartorius BIA Separations, Ajdovscina, Slovenia

Mesenchymal stem cell derived extracellular vesicles have promising therapeutic potential in diverse applications, such as tissue regeneration and immunomodulation. On the pathway to the clinic, there is a substantial need for development of scalable methods for EV production and isolation. Moreover, advancements in analytical tools for characterization of EVs are necessary to ensure a consistent product. Here, we showcase an integrative approach combining upstream, downstream and analytical processes for successful generation of MSC EVs.

This poster is presented by Simon Staubach (Sartorius).

Abstract No. P137

Exploring cardiac repair mechanisms with isolated and immortalized epicardium-derived cells

Katja Nowak[1,2,*], *René Schramm*[1], *Barbara Kaltschmidt*[2], *Christian Kaltschmidt*[2], *Cornelius Knabbe*[1], and *Anna L. Höving*[1,2]

[1] Heart and Diabetes Centre NRW, Bad Oeynhausen
[2] University of Bielefeld

*Presenting author

The epicardium is a key contributor to cardiac development, providing various cell types via epithelial-to-mesenchymal transition (EMT) and secreting paracrine factors that promote cardiac tissue formation. While it remains quiescent in a healthy adult heart, ischemic injuries such as myocardial infarction can stimulate its reactivation and support repair processes. Epicardium-derived cells (EPDCs) isolated from the adult human heart auricle present a promising opportunity to explore the epicardium's role in heart regeneration. We successfully isolated primary EPDCs from the human left atrial appendage and described their marker profile and potential for EMT in culture. Furthermore, the cells were immortalized using SV40LT and show expression of various markers associated with the epicardium and EPDCs. To understand their potential role in cardiac repair, further studies are initiated to examine EMT and migration in EPDCs.

Abstract No. P138

Optimizing pluripotent stem cell spheroid growth in liter scale bioreactors through the use of constant medium perfusion

Michael Akenhead, *Mitchell Maloy*, *Marcus Bunn*, *Abigail Becker*, *Navjot Kaur*, *Evan Zynda*, *Mark Kennedy*, *Sara Perez Munoz**, and *David Kunninger*

ThermoFisher Scientific, Frederick, US & Madrid, Spain

*Presenting author

In order to efficiently meet clinical manufacturing needs requiring high quantities of pluripotent stem cells (PSCs), the development and optimization of PSC culture methods in large volume bioreactors is critical. We have previously shown how PSCs can be expanded as spheroids in 3L stirred-tank reactors (STRs). These bioreactors use impellers that continuously mix the medium but also generate shear stress that may negatively affect PSC growth. Additionally, current protocols that rely on gravity sedimentation of PSC spheroids and manual aspiration of spent medium are impractical beyond a 3L culture scale. Notably, STRs can be combined with perfusion systems to constantly supply fresh medium and mitigate the need to manually exchange medium. Here, we describe our efforts to optimize PSC spheroid growth in liter scale bioreactors by balancing stir speeds, shear stress effects, and medium exchange rates using constant perfusion of StemScale PSC Suspension Medium. Cells were initially seeded into STRs at low stir speeds to promote spheroid formation. The speed was then gradually increased to prevent spheroid aggregation and maintain culture homogeneity. Shear was detrimental to spheroid growth at high speeds, though this could be minimized with shear protectants. Our results indicated that the addition of shear protectant enabled spheroids to grow larger and more uniform in size, resulting in greater cell yields. We further evaluated spheroid growth by using tangential flow depth filtration (TFDF) and alternating tangential flow (ATF) perfusion systems to perform medium exchanges. We determined that both systems were able to support spheroid expansion with similar efficiencies. Taken together, with these optimized parameters we observed yields of up to 20 billion PSCs (10x106 cells/mL, equaling ~80-fold expansion) capable of maintaining pluripotency (>95% OCT4+/NANOG+ cells) after 10 days in culture. Overall, these results demonstrate PSC culture in perfused STRs is an effective means to improve PSC expansion workflows.

Abstract No. P139

Off the shelf immune-shielded stem cell-derived γδ T cells for clinical applications

Tommaso Pezzica[1,*], *Stephanie Sontag*[1], *Iva Tjoeng*[1], *Judith Hartmann*[1], *Stefanie Kober*[1], *Lea Renneberg*[1], *Jessica Nolte*[1], *Theresa Klisch*[1], *Thomas Aleyt*[1], *Caroline Stienne*[2], *Philip Hublitz*[1], *Michael Esquerre*[2], *Katrin Ridders*[1], *Matthias Austen*[1], and *Audrey Holtzinger*[1]

[1] Cell Therapy, Evotec International GmbH, Göttingen
[2] Immuno-Oncology, In Vitro Biology, Evotec SE, Toulouse, France

*Presenting author

Chimeric antigen receptor (CAR) T cell immunotherapies have revolutionized medicine and were successfully used to treat hematological and solid malignancies, infectious and autoimmune diseases. While αβ CAR T cells have shown good efficacy, they have also raised safety concerns e.g. due to life-threatening graft versus host disease. In contrast, γδ CAR T cells were shown to be safe but having only moderate efficacy likely because unedited γδ T cells have a low target specificity and are quickly rejected. Genetic engineering of induced pluripotent stem cells (iPSCs) e.g. by introducing a CAR or immune evasion gene edits offers an indefinite and homogenous starting material for the *in vitro* differentiation of iPSC-derived γδ T cells (iγδ T cells) as therapeutic product of defined genotype, phenotype and purity.

Here, we report on i) generating GMP-line-derived immune-shielded iPSCs that serve as platform for customized CAR iγδ T cell therapies and ii) a scalable xeno-free *in vitro* differentiation process allowing for the large-scale production of immune-shielded iγδ T cells for clinical applications.

Engineered immune-shielded iPSCs are differentiated into iPSC-derived CD34+ (iCD34+) cells with a bona fide hematopoietic progenitor phenotype. As the CD34 purity achieved with our protocol is high, hematopoietic progenitor cells can be further differentiated into iγδ T cells without any CD34 enrichment step. iγδ T cells are differentiated and expanded in a feeder- and serum-free environment reaching a high purity and yield. Upon activation, iγδ T cells are functional and display a naïve, non-exhausted, immune-shielded phenotype.

The combination of efficient manufacturing with targeted genetic modifications results in an iγδT cell platform that can be rapidly adapted for the treatment of different diseases e.g. in oncology or autoimmunity.

Abstract No. P140

Clinical scale non-viral gene editing and feeder-free production of CAR-NK cells

Deepak Kumar[1], *Olga Cohen*[1], *Sung Lee*[1], *Ranganatha R Somasagara*[1], *Pushpalatha Chaluvappa*[1], *Greogory Bonello*[1], *Corinna Pohl*[2,*], and *Namritha Ravinder*[1]

[1] Thermo Fisher Scientific, Carlsbad, US
[2] Thermo Fisher Scientific, Darmstadt

*Presenting author

Purpose: The development of clinically relevant and efficient, non-viral gene delivery methods for primary NK cells remains limited. In this study, we optimized a scalable, non-viral, gene delivery method for production of CAR-NK cells.

Methods: NK cells were isolated from PBMC using Gibco CTS Rotea counterflow centrifugation system and cultured in NK expander complete media supplemented with IL-2 for 6 days. On day 6, NK cells were electroporated with Neon NxT or Gibco CTS Xenon electroporation system at density of 50e06 cells/mL to deliver single strand DNA (ss-DNA), CRISPR-Cas9 protein and synthetic gRNA targeting Rab11a/AAVS1 loci. The gene edited NK-cells were expanded and analyzed by flow cytometry for CAR expression. Cells were also further tested for cytotoxicity by co-culturing with SKOV3-GFP for 6 hrs.

Results: GFP knock-in at Rab11a locus demonstrated up to 25% knock-in efficiency while ant-meso-3 CAR knock-in at AAVS1 locus was up to 15% over the time of culture. The edited NK cells maintained their phenotype and viability. Furthermore, engineered NK cells (anti-meso-3 CAR NK) showed higher cytotoxicity effect towards target cancer cells as compared to non-engineered NK cells.

Abstract No. P141

Clonal and lineage tracing of barrier mesothelium

Aydan Sardogan[1,2,]*, *Yiqun Su[1]*, *Donovan Correa-Gallegos[3]*, *Tim Koopmans[1,4]*, *Andy Qarri[1]*, *Tingting Ling[5]*, *Ashesh Chakraborty[2]*, *Bikram Dasgupta[1]*, *Haifeng Ye[1, 6]*, *Christoph Desienhofer[1]*, *Subhasree Dutta[1,2]*, *Jiakuan Zhao[1]*, *Marie-Christin Weber[7]*, *Ali Önder Yildirim[2,8]*, *Carsten Marr[9, 10]*, and *Yuval Rinkevich[1,5]*

[1] Institute of Regeneration Biology and Medicine, Helmholtz Munich
[2] Institute of Lung Health and Immunity, Helmholtz Munich
[3] Institute for Stroke and Dementia, Ludwig-Maximilians University Hospital, Munich
[4] Hubrecht Inst., KNAW & Uni. Medical Center Utrecht, The Netherlands
[5] Inst. Reg. Biol. & Med., Chinese Inst. Med. Res. & Capital Med. Uni., Beijing, China
[6] Med. Center Burn Plastic and Wound Repair, Jiangxi Med. Coll., Nanchang Uni., China
[7] Dep. Surgery, TUM School of Medicine, Rechts der Isar Hosp., Technical Uni. of Munich
[8] Inst. of Exp. Pneumology, LMU Uni. Hosp., Ludwig-Maximilians Uni., Munich
[9] Institute of AI for Health, Helmholtz Munich
[10] Artificial Intelligence in Hematology and Cell Therapy, LMU Uni. Hosp., Munich

*Presenting author

The mesothelium is the largest continuous epithelial monolayer that lines the surfaces of internal organs and their associated cavities. Mesothelial cells derived from the lateral plate mesoderm, preserve serosal integrity by secreting inflammatory mediators, growth factors, and extracellular matrix components. These cells display both epithelial and mesenchymal features with high repair potential. Under homeostatic conditions, mesothelial cells exhibit low proliferative activity; however, upon injury, they undergo rapid proliferation and mesothelial-to-mesenchymal transition to restore epithelial integrity. Beyond their reparative role, mesothelial cells also contribute to pathological processes such as postsurgical adhesions and idiopathic pulmonary fibrosis. Previous studies have predominantly relied on transgenic mouse models (e.g., MSLN-CreER, WT1-CreER) to trace mesothelial lineages in adulthood. However, these models often lack specificity, inaccurately labeling other cell types and tissues, and therefore are limited in their ability to investigate mesothelial development and function. To overcome these limitations, we used two inducible CreER knock-in reporter mouse lines (Procr-CreER and PDPN-CreER) and performed single-cell lineage tracing, clonal analysis, cell division, and single-cell RNA sequencing to investigate mesothelial dynamics during mouse postnatal development, adult, and injury models including lung fibrosis, adhesions, and lipopolysaccharide-induced inflammation. Both Procr and PDPN inducible lines revealed significant increase in clonal expansion under postnatal and injury conditions compared to the adult stage. We further investigated the role of the basement membrane in regulating mesothelial cell behaviour. Basement membrane was associated with cell clonality and proliferation in both postnatal development and aforementioned injury conditions. Our study sheds light on the mechanisms of mesothelial proliferation, cell-cell and cell-matrix interactions, and tissue repair, thus providing valuable insights for therapeutic exploration in pathological conditions such as fibrosis and infection.

Abstract No. P142

Retrospective gene expression analysis in neural stem cells

(from "Stem cells in aging & disease (cancer stem cells)")

Zeynep Aydin[1,*], *Svetlana Ovchinnikova*[2], *Anna Martin-Villalba*[1], and *Simon Anders*[2]

[1] Molecular Neurobiology, German Cancer Research Center (DKFZ), Heidelberg
[2] BioQuant, Heidelberg University

*Presenting author

Understanding how individual stem cells transition between distinct states is fundamental to studying development, regeneration, and disease. While single-cell RNA sequencing (scRNA-seq) has revealed the diversity of cellular states within tissues, it mainly provides static snapshots, and computational predictions are required to infer cell trajectories. Traditional lineage-tracing approaches enable clonal fate mapping but lack the resolution to capture the dynamic transcriptional history of individual cells.

To overcome these limitations, we developed the DCM Time Machine (DCM-TM), an experimental tool that links a cell's past and present by combining single-cell transcriptome and DNA methylation profiling and a bacterial DCM methyltransferase. Subsequent single-cell nucleosome methylome and transcriptome sequencing (scNMT-seq) enables simultaneous reconstruction of past transcriptional activity and current cellular states.

We established proof-of-concept for DCM-TM, demonstrating that DCM methylation patterns accurately reflect transcriptional profiles and allow robust discrimination of distinct cell types and subpopulations. This method provides a powerful framework for studying stem cell fate decisions, enabling the reconstruction of transcriptional trajectories during differentiation, tissue regeneration, and responses to injury.

By directly linking historical gene activity with current cell identity, DCM-TM opens new avenues for understanding how stem cells maintain plasticity, commit to specific lineages, and respond to environmental cues, insights essential for developmental biology, regenerative medicine, and stem cell-based therapies.

Abstract No. P143

ELAPOR2 regulates proliferation of pancreatic progenitors and endocrine differentiation

(from “Pluripotency, differentiation & PSC-derived models”)

Osvaldo Rodrigues Pereira[1,*], *Johanna Siehler*[1,2], *Kei Kozawa*[1], *Ingo Burtscher*[1], and *Heiko Lickert*[1]

[1] Helmholtz Diabetes Center, Helmholtz Munich
[2] Vertex Pharmaceuticals, Boston, US

*Presenting author

The pancreas is an essential organ for endocrine regulation of glucose metabolism, and its proper development depends on tightly coordinated genetic and cellular programs.

Pancreatic progenitor specification, branching morphogenesis, and the balance between endocrine and exocrine lineages all require precise molecular control, and perturbations in these processes can have long-lasting effects on organ function.

Previous studies have identified ELAPOR2 as a putative regulator of proliferation throughout development, expressed throughout all germ layers, but its specific role in the pancreatic endoderm remains poorly understood.

Using stem cell differentiation as a model of pancreas development and endocrine differentiation, we show that loss of ELAPOR2 directly affects pancreatic progenitor expansion, increasing the proportion of progenitors going through the cell cycle, and influences endocrine differentiation, enhancing the yield of β-cells.

These findings reveal that ELAPOR2 is an active regulator of progenitor expansion and cell fate. Our results add to previous knowledge by identifying a new molecular player in the regulatory network governing pancreas formation, and a potential target for differentiation towards the β-cell fate.

Abstract No. P144

Extraction by AI for tracking of hPSC-based clinical trials

Meiying Xue[1,*], *Nancy Mah*[2], *Sabine Kobold*[2], and *Andreas Kurtz*[1]

[1] Charité Universitätsmedizin Berlin, BIH Center for Regenerative Therapies, Berlin
[2] Biomedical Data and Bioethics Group, Fraunhofer Institute for Biomedical Engineering, Sulzbach

*Presenting author

With the growing number of clinical studies involving human pluripotent stem cell (hPSC)-derived products, the human pluripotent stem cell registry (www.hpscreg.eu) clinical database has become a key infrastructure in the field. However, data curation remains largely manual, posing challenges in terms of scalability and timely updates. To address this, we developed a natural language processing (NLP)-based automated pipeline to identify and classify clinical trials related to induced pluripotent stem cells (iPSCs) and embryonic stem cells (ESCs).

Our system integrates over one million clinical trial records from multiple trial registries and applies semantic normalization using biomedical ontologies such as MeSH and the Cell Ontology. Key fields—such as study title, intervention type, disease indication, trial phase, and product name—are extracted and standardized. Semantic embeddings derived from a domain-general biomedical language model are used in combination with a support vector machine (SVM) classifier to predict trial relevance. To improve robustness, we optimized feature extraction strategies, adapted data preprocessing pipelines, and addressed label imbalance. Additionally, a curated company–product mapping was developed to support the inference of stem cell relevance in studies lacking explicit terminology.

The system has been deployed to assist with structured data updates for the hPSCreg database and serves as a foundation for future visualization and API development. This work demonstrates the feasibility of applying language model technologies to biomedical database curation, supporting transparency and traceability in stem cell clinical research beyond PSC.

Author Index

A

Abitorabi, Karin C14
Afonso Martins, Inês P052
Aicher, Jasmin T33
Al Hijailan, Reem P007
Albert, Mareike T10
Alptekin, Sena T39
Annunziata, Chiara P082
Apáti, Ágota P083
Artioli, Annasara P008
Austen, Matthias C11
Aydin, Zeynep P142

B

Bageritz, Josephine P048
Baier, Eleonore T48
Benkisser-Petersen, Marco C08, P009
Berger, Constantin P010
Berger, Thomas T02
Beyer, Caroline P066
Bhagat, Pranjali Milind P011
Bhamidipati, Ankita Kumar P084
Binder, Stephanie P085
Blank-Stein, Nelli P086
Blenkle, Alica T31
Boccellato, Francesco P053
Boder, Philipp P012
Boolani, Adil P054
Brito, Catarina S3
Brownlee, Robert C03
Bulut-Karslioglu, Aydan PS2
Büschel, Ida P073

C

Calzolari, Filippo P125
Chan, Andy Shing-Fung T53
Chan, Kazette Yuen Yu P087
Cicek, Ekin Gökce P013

D

Dannert, Angelika P088
Debowski, Katharina C02
Dobner, Jochen T27
Duda, K. Julia C10
Duffy, Amy P014

E

Ellinger, Tobias T09
Es Sbai, Saloia P089
Esk, Christopher P056

F

Farkas, Falk Janos T38
Felski, Christian P090
Finke, Marcel P001
Firuzi, Rezwan P126
Frye, Michaela PS3
Fröbel, Julia T17
Funk, Maja T36

G

Gasparotto, Matteo T34
Glaser, Markus P092
Glover, Joel C. S6
Godthardt, Kathrin P015
Gohl, Thomas P093
Golda, Gosia T08
Graffmann, Nina P017
Greber, Boris P127
Greisle, Tobias T45
Gross-Thebing, Theresa T29
Guida, Catello P058
Guledani, Aleks P094
Günther, Katharina P128

H

Haasen, Nele S5
Haberhausen, Diana P018, T04
Hafner, Leon P074
Hahnau-Jurth, Sabine C05
Hamad, Sarkawt P059
Hanna, Jacob K3
Hauser, Stefan P095
Heiduschka, Sonja P096
Henning, Nina P049
Hernandez Bautista, Carlos A. P019
Herrmann, Sabrina P020
Hinkelmann, Alica P130
Hofmann, Samuel P097
Hohwieler, Meike P021, P098
Hoja, Matthias C09
Horland, Reyk P099
Horlova, Anastasiia P075
Horschitz, Sandra P100
Hradil, Vojtech P131
Hufschmid, Katja P132
Hörmanseder, Eva T50

I - J

Inaas, Syeda P022
Jin, Qianren P024
Jolly, Adrien P076
Jung, Jessica P102
Jung-Klawitter, Sabine P103

K

Kafadar, Selen P002
Karampelias, Christos P060
Karnatz, Isabell P104
Kaysan, Leon P061
Ketteler, Carolin P105
Keßler, Antonia P026
Khan, Sohana Islam P027
Kipper, Sophie P133
Kleemiß, Felix P106
Klein, Dominik P077
Koch, Philipp P107
Koufali, Angeliki P062
Krutenko, Tamara P108
Kuleshova, Anna P028
Külp, Marius T18

L

Lang, Sarah P029
Lansche, Christine P031
Laufer, Sandra D. P109
Lemme, Marta P110
Liang, Manqi P030
Lithovius, Väinö T05
Lu, Ziwei P032
Luo, Yanyan P111

M

Mansour, Gizem P033
Marcato, Silvia P112
Markova, Pavlina P063
Martin-Villalba, Ana T06
Maurer, Jochen P050
Metzger, Jakob S1
Minaeva, Mariia T52
Münchhalfen, Lisa P134

N

Nagy, Andras T25
Nebol, Aylin P113
Niemöller, Theresa P135
Nischalke, Vera P036
Nold, Philipp C01
Nowak, Katja P137
Nugraha, Osman Abhimata P078
Nurieva, Wasifa P114

O

Özata, Gamze P115
Ozaki, Mari T37

P - Q

Panza, Paolo T42
Paquet, Dominik T32
Perez Munoz, Sara P138
Pezzica, Tommaso P139
Pfaender, Stefanie T03
Pförtner, Felix P079
Piazzalonga, Francesca P037
Plötner, Clara Luise P116
Podlesnic, Martina P117
Pohl, Corinna P140
Puertollano Martín, David P118
Puri, Deepika T47
Pérez Fernández, Raquel P064
Quach, Susanna P065

R

Rafiei Hashtchin, Anna P066
Reigl, Amelie T13
Reinelt, Anna T14
Reinhardt, Peter S4
Risse, Isabelle P038
Rodrigues Pereira, Osvaldo P143
Romeike, Merrit P119
Romero-Mulero, Mari Carmen T19
Rädecke, Kristin P039

S

Saleh, Fawaz P003
Sardogan, Aydan P141
Scarcella, Daniele P080
Schaller, Heidrun P004
Scheinpflug, Philine P130
Schlichthärle, Thomas T12
Schmachtel, Tessa P051
Schmidt, Aileen T41
Schmidt, Malin P120
Schmoll, Kea Aline P121
Schroeder, Timm K2
Schröter, Christian T46
Schumann, Gerald G. S8
Schupp, Jonas T44
Schurer, Amelie P067
Shelashen, Arina T51
Shigematsu, Kazuki P040, P068
Spathopoulou, Angeliki T26
Srinidhi, Jahnavi P041
Stewart, Liz C13
Stock, Marco P081
Stricker, Stefan T07

T

Torres-Padilla, Maria-Elena K5
Teichert, Ferdinand P042
Terheyden-Keighley, Daniel P005
Tian, Chenglei P043, P044
Tolle, Isabella P122
Trpchevska, Ana P070

U - V

Uxa, Sigrid T20
Vincendeau, Michelle P045

W

Wasnick, Roxana Maria P071
Weller, Arjen P123
Wessel, Tim P006
Wilbertz, Johannes S2
Wilkinson, Adam T16
Wilson, Liam T49
Wörner, Nils P046

Z

Zeevaert, Kira	T28
Zenagui, Reda	C12
Zimmer, Bastian	P124, T30
Zink, Annika	T15

List of participants

A

Aberoumandi, Mohsen
MedChemExpress, Sollentuna, SE
mohsen.aberoumandi[at]medchemexpress.com

Abitorabi, Karin
Minaris Advanced Therapies, Ottobrunn
k.abitorabi[at]rm.minaris.de

Abou Khalil, Nour
Stem Genomics, Grabels, FR
annick.marcellin[at]stemgenomics.com

Aicher, Jasmin
Institute of Reconstructive Neurobiology, Bonn
jaicher[at]uni-bonn.de

Al Hejailan, Reem
King Faisal Specialist Hospital and Reserach Center, Riyadh, SA
rhijailan[at]kfshrc.edu.sa

Albert, Mareike
Technische Universität Dresden, Dresden
mareike.albert[at]tu-dresden.de

Alhashimi, Fatma
First Stem Cell & Genomics Laboratory, Dubai, AE
fatma.alhashimi[at]fsg-lab.com

Alptekin, Sena
Deutsches Krebsforschungszentrum (DKFZ), Heidelberg
senaalptekin[at]hotmail.com

AlShaer, Mohammad
King Saud University, Riyadh, SA
dr.mohammadalshaier[at]gmail.com

Amemiya, Seika
PeptiGrowth, Inc., Tokyo, JP
seika.amemiya[at]peptigrowth.com

Anastassiadis, Konstantinos
BIOTEC, TUD, Dresden
konstantinos.anastassiadis[at]tu-dresden.de

Annunziata, Chiara
BioLamina, Sundbyberg, SE
chiara.annunziata[at]biolamina.com

Apati, Agota
HUN-REN Research Centre for Natural Sciences, Budapest, HU
apati.agota[at]ttk.hu

Arps, Volker
Bio-Techne, Wiesbaden
volker.arps[at]bio-techne.com

Artioli, Annasara
Central Institute for Mental Health, Mannheim
annasara.artioli[at]zi-mannheim.de

Askurava, Tamari
Meat Tomorrow, Skovlunde,DK
taas[at]meattomorrow.com

Austen, Matthias
Evotec, Goettingen
matthias.austen[at]evotec.com

Aydin, Zeynep
German Cancer Research Center (DKFZ), Heidelberg
zaydin1994[at]gmail.com

B

Babl, Leon
Thermo Fisher Scientific, Darmstadt
leon.babl[at]thermofisher.com

Baddack-Werncke, Uta
DLR Projektträger, Bonn
uta.baddack-werncke[at]dlr.de

Bageritz, Josephine
Heidelberg University, Heidelberg
josephine.bageritz[at]cos.uni-heidelberg.de

Baier, Eleonore
TUM Klinikum Rechts der Isar, Technical University of Munich, Munich
eleonore.baier[at]tum.de

Bakhti, Mostafa
Institute of Diabetes and Regeneration Research, Neuherberg
mostafa.bakhti[at]helmholtz-munich.de

Baptista, Patricia
Stem Genomics, Grabels, FR
patricia.baptista[at]stemgenomics.com

Bartfeld, Sina
Technical University (TU) Berlin, Berlin
s.bartfeld[at]tu-berlin.de

Bartscherer, Kerstin
Osanbrück University, Osnabrück
kerstin.bartscherer[at]uni-osnabrueck.de

Batool, Lilas
Human Center for Kidney Health, UKE Hamburg, Hamburg
l.batool[at]uke.de

Baumann, Jacob
Institut für Pharmazeutische Biotechnologie, Biberach
jacob.baumann[at]uni-ulm.de

Baumgart, Emily Violette
Institute of Diabetes and Regeneration Research, Neuherberg
emilyviolette.baumgart[at]helmholtz-munich.de

Bayer, Dennis
Zymo Research Europe, Freiburg
dbayer[at]zymoresearch.de

Benkisser-Petersen, Marco
Sartorius Lab Instruments GmbH & Co. KG, Göttingen
marco.benkisser-petersen[at]sartorius.com

Berger, Constantin
Fraunhofer ISC - TLZ, Würzburg
constantin.berger[at]isc.fraunhofer.de

Berger, Thomas
Catalent Düsseldorf, Langenfeld
thomas.berger[at]catalent.com

Besser, Daniel
German Stem Cell Network (GSCN), Berlin
daniel.besser[at]bih-charite.de

Bhagat, Pranjali Milind
Uniklinik Köln, Cologne
pranjalibhagat2020[at]gmail.com

Bhamidipati, Ankita Kumar
Zentralinstitut für Seelische Gesundheit, Mannheim
ankita.kumar[at]zi-mannheim.de

Bhattarai, Marcel
MACHEREY-NAGEL GmbH & Co. KG, Düren
mbhattarai[at]mn-net.com

Binder, Stephanie
IUF - Leibniz Research Institue for Environmental Medicine, Düsseldorf
stephanie.binder[at]iuf-duesseldorf.de

Blank-Stein, Nelli
Life & Medical Sciences Institute, University of Bonn, Bonn
nelli.blank[at]uni-bonn.de

Blaszkiewicz, Sandra
Becton Dickinson GmbH, Heidelberg
sandra.blaszkiewicz[at]bd.com

Bleischwitz, Marc
Integrated DNA Technologies, Munich
mbleischwitz[at]idtdna.com

Blenkle, Alica
Fraunhofer Institute for Translational Medicine and Pharmacology ITMP, Translational Neuroinflammation and Automated Microscopy TNM, Göttingen
alica.blenkle[at]itmp.fraunhofer.de

Boder, Philipp
Amsbio, Alkmaar, NL
philippb[at]amsbio.com

Böhm, Christiana
HAMILTON Germany GmbH, Rosenheim
Cboehm[at]hamilton.ch

Böhmerova, Eva
European Patent Office, Munich
ebohmerova[at]yahoo.de

Boolani, Adil
European Molecular Biology Laboratory Heidelberg, Heidelberg
boolani.adil[at]gmail.com

Borisova, Elitsa
Julius-Maximilians-Universität Würzburg/Catalent Düsseldorf, Cologne
elitsa.borisova[at]stud-mail.uni-wuerzburg.de

Böttcher, Anika
Helmholtz Munich, Neuherberg
anika.boettcher[at]helmholtz-munich.de

Bracht, Stephanie
Biozym Scientific GmbH, Hess. Oldendorf
stephanie.bracht[at]biozym.com

Brachthäuser, Maite
Fraunhofer ITMP, Hamburg
maite.brachthaeuser[at]ruhr-uni-bochum.de

Braun, Thomas
Max Planck Institute for Heart and Lung Research, Bad Nauheim
office.braun[at]mpi-bn.mpg.de

Brimpari, Minodora
Institute of Translational Stem Cell Research, Oberschleißheim
minodora.brimpari[at]helmholtz-munich.de

Brito, Catarina
ITQB NOVA & iBET, Oeiras, PT
anabrito[at]ibet.pt

Brownlee, Robert
Opes Diagonostics, Market Harborough, UK
robert[at]opesdiagnostics.com

Brüstle, Oliver
Bonn University, Bonn
oliver.bruestle[at]uni-bonn.de

Bukas, Christina
Helmholtz AI, Berlin
christina.bukas[at]helmholtz-munich.de

Bulut-Karslioglu, Aydan
Max Planck Institute for Molecular Genetics, Berlin
aydan.karslioglu[at]molgen.mpg.de

Büschel, Ida
LMU/Helmholtz Munich, Munich
ida.bueschel[at]gmail.com

C

Cabezas Wallscheid, Nina
Max Planck Institute for Immunobiology and Epigenetics, Freiburg
cabezas[at]ie-freiburg.mpg.de

Calzolari, Filippo
JGU, Institute for Physiological Chemistry, Mainz
fcalzola[at]uni-mainz.de

Camargo, German
ETH Zürich, Basel
german.camargo[at]bsse.ethz.ch

Cantz, Tobias
Hannover Medical School, Hannover
cantz.tobias[at]mh-hannover.de

Cayir, Sefa Berkay
Helmholtz Munich, Neuherberg
sefaberkay.cayir[at]helmholtz-munich.de

Chan, Andy Shing-Fung
Würzburg Institute of Systems Immunology, Würzburg
shing-fung.chan[at]uni-wuerzburg.de

Chan, Kazette Yuen Yu
University of Alberta, Edmonton, CA
kazettey[at]ualberta.ca

Cicek, Ekin Gokce
Institute of Diabetes and Regeneration, Helmholtz Munich, Munich
ekin.cicek[at]helmholtz-munich.de

Cornily, Victor
Universität Osnabrück, Osnabrück
victor.cornily[at]uni-osnabrueck.de

Cozzitorto, Corinna
Helmholtz Munich, Neuherberg
corinna.cozzitorto[at]helmholtz-munich.de

Cross, Michael
University of Leipzig, Leipzig
crossm[at]medizin.uni-leipzig.de

Cyrys, Kevin
Lebao, MHH, Hannover
ullmann.kevin.httg[at]mh-hannover.de

D

Dannert, Angelika
University Hospital LMU Munich, Munich
angelika.dannert[at]med.uni-muenchen.de

de Heer, Arnold
ADS Biotec, Glasgow, UK
adeheer[at]adsbiotec.com

Debowski, Katharina
STEMCELL Technologies, Cologne
katharina.debowski[at]stemcell.com

Deng, Hongkui
Peking University, Bejing, CN
hongkui_deng[at]pku.edu.cn

Diecke, Sebastian
Max Delbrück Center for Molecular Medicine, Berlin
sebastian.diecke[at]mdc-berlin.de

Dobner, Jochen
IUF - Leibniz Research Institute for Environmental Medicine, Düsseldorf
jochen.dobner[at]iuf-duesseldorf.de

Dowbaj, Anna
TUM Center for Organoid Systems, Garching
anna.dowbaj[at]tum.de

Drakhlis, Lika
Hannover Medical School, Hannover
drakhlis.lika[at]mh-hannover.de

Drobny, Alice
Mettler Toledo GmbH, Gilching
alice.drobny[at]mt.com

Drukker, Micha
Leiden Academic Centre for Drug Research (LACDR), Leiden, NL
m.drukker[at]lacdr.leidenuniv.nl

Duda, K. Julia
Integrated DNA Technologies, Munich
kduda[at]idtdna.com

Duffy, Amy
Royal College of Surgeons in Ireland, Beaumont, IE
amyduffy23[at]rcsi.com

E

Edenhofer, Fiona
LMU Munich, Munich
edenhofer[at]bio.lmu.de

Edenhofer, Frank
Molecular Biology, Innsbruck, AT
frank.edenhofer[at]uibk.ac.at

Eiberger, Jürgen
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
juergene[at]miltenyibiotec.de

Eichwald, Sabrina
Leibniz Institute on Aging Research, Jena
sabrina.eichwald[at]leibniz-fli.de

Elanzew, Andreas
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
andreasel[at]miltenyibiotec.de

Ellinger, Tobias
Klinikum rechts der Isar, Dorfen
tobias.ellinger[at]tum.de

Elster, Anda
Proteintech, Planegg-Martinsried
anda[at]ptglab.com

Engel, Peter
OMNI Life Science GmbH & Co. KG, Bremen
peter.engel[at]ols-bio.de

Erserim, Inanc Deger
Biomerieux, Nürtingen
inanc.erserim[at]biomerieux.com

Es Sbai, Saloia
University Hospital Bonn, Bonn
saloia.es[at]smail.bcw.h-brs.de

Esk, Christopher
Institute of. Molecular Biology, Innsbruck, AT
christopher.esk[at]uibk.ac.at

Essers, Marieke
HI-STEM gGmbH, Heidelberg
marieke.essers[at]hi-stem.de

F

Farkas, Falk
Institute of Diabetes and Regeneration, Munich
falk.farkas[at]helmholtz-munich.de

Felski, Christian
Institute of Reconstructive Neurobiology, Bonn
cfelski[at]uni-bonn.de

Feng, Jin
Institute of Diabetes and Regeneration Research, Neuherberg
jin.feng[at]helmholtz-munich.de

Finazzi, Veronica
Helmholtz Munich, Planegg
veronica.finazzi[at]helmholtz-munich.de

Finke, Marcel
BIH Center for Regenerative Therapies at Charité Berlin, Berlin
marcel.finke[at]bih-charite.de

Firuzi, Rezwan
The Marga and Walter Boll Laboratory for Cardiac Tissue Engineering, Cologne
rezwanfiruzi[at]gmail.com

Fischer, Benjamin
Evotec, Hamburg
benjamin.fischer[at]evotec.com

Fischer, Iris
National Network Office GCT Germany, Berlin
iris.fischer2[at]bih-charite.de

Fischer, Joachim
ADS Biotec, Glasgow, UK
jfischer[at]adsbiotec.com

Frahm, Silke
Max Delbrück Center, Berlin
sfrahm[at]mdc-berlin.de

Freund, Christian
Leiden University Medical Center, Leiden, NL
c.m.a.h.freund[at]lumc.nl

Fröbel, Julia
Leibniz Institute on Aging – Fritz Lipmann Institute (FLI), Jena
julia.froebel[at]leibniz-fli.de

Frömer, Andreas
Biomol GmbH, Hamburg
a_froemer[at]biomol.de

Froriep, Gaby
REBIRTH, Hannover
froriep.gaby[at]mh-hannover.de

Frye, Michaela
German Cancer Research Center - DKFZ, Heidelberg
m.frye[at]dkfz.de

Funk, Maja
Helmholtz Munich, Neuherberg
maja.funk[at]helmholtz-munich.de

Fürst, Anna
ISAR Bioscience GmbH, Planegg
anna.fuerst[at]isarbioscience.de

G

Ganguly, Archisha
University Medical Centre, Göttingen, Göttingen
archisha.ganguly[at]med.uni-goettingen.de

Gasparotto, Matteo
Central Institute for Mental Health (CIMH), Mannheim
matteo.gasparotto[at]zi-mannheim.de

Geiger, Hartmut
Ulm University, Ulm
hartmut.geiger[at]uni-ulm.de

Ghodsi, Saman
Osnabrück University, Osnabrück
saman.ghodsi[at]uni-osnabrueck.de

Glaser, Markus
University Hospital Würzburg, Würzburg
markus.glaser[at]uni-wuerzburg.de

Glauche, Ingmar
Technische Universität Dresden, Dresden
ingmar.glauche[at]tu-dresden.de

Glover, Joel
University of Olso, Norwegian Stem Cell Center, Oslo, NO
j.c.glover[at]medisin.uio.no

Godthardt, Kathrin
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
kathring[at]miltenyi.com

Gohl, Thomas
Telight, Hallbergmoos
thomas.gohl[at]telight.eu

Golda, Gosia
Würzburg Institute of Systems Immunology/Max Planck Institute of Immunobiology and Epigenetics, Würzburg
golda[at]ie-freiburg.mpg.de

Götze, Susanne
ISAR Bioscience GmbH, Planegg
susanne.goetze[at]isarbioscience.de

Gouti, Mina
Max Delbrück Center for Molecular Medicine, Berlin
mina.gouti[at]mdc-berlin.de

Graffmann, Nina
Medical Faculty and University Hospital Düsseldorf, Heinrich Heine University, Düsseldorf, Ratingen
nina.graffmann[at]med.uni-duesseldorf.de

Grapin-Botton, Anne
Max Planck Institute for Molecular Cell Biology and Genetics, Dresden
anne.grapin-botton[at]mpi-cbg.de

Greber, Boris
Catalent Düsseldorf GmbH, Langenfeld
boris.greber[at]catalent.com

Greif, Gabriela
European Patent Office, Munich
gabrielagreif[at]europe.com

Greisle, Tobias
Helmholtz Munich, Munich
tobias.greisle[at]helmholtz-munich.de

Gross-Thebing, Theresa
Gurdon Institute, University of Cambridge, Cambridge, UK
tg477[at]cam.ac.uk

Gross, Christian
Thermo Fisher Scientific, Courtaboeuf (Villebon-sur-Yvette), FR
christian.gross[at]thermofisher.com

Grün, Dominic
Institute of Systems Immunology, Würzburg
dominic.gruen[at]uni-wuerzburg.de

Guezguez, Borhane
University Medical Center Mainz, Mainz
guezguez[at]uni-mainz.de

Guida, Catello
Heidelberg University, Mannheim
catello.guida[at]zi-mannheim.de

Guledani, Aleks
SYNENTEC GmbH, Elmshorn
aguledani[at]synentec.com

Günther, Katharina
Green Elephant Biotech, Berlin
events[at]greenebt.com

H

Haas, Simon
Berlin Institute of Medical Systems Biology, Berlin
simon.haas[at]mdc-berlin.de

Haasen, Nele
konteam, Füssen
haasen[at]konteam.de

Haberhausen, Diana
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
dianaha[at]miltenyi.com

Hachani, Eymen
FUJIFILM Wako Chemicals, Neuss
eymen.hachani[at]fujifilm.com

Hafner, Leon
Institute of Computational Biology, Erding
leon.hafner[at]helmholtz-munich.de

Hahnau-Jurth, Sabine
Biomerieux, Nürtingen
sabine.hahnau[at]biomerieux.com

Hamad, Sarkawt
Marga-and-Walter-Boll Laboratory, Cologne
shamad[at]uni-koeln.de

Hamanaka, Shiro
IVICT Europe GmbH, Düsseldorf
shiro.hamanaka[at]ivic-t.com

Hanke, Miriam
New England Biolabs GmbH, Frankfurt am Main
hanke[at]neb.com

Hanna, Jacob
The Weizmann Institute of Science, Rehovot
jacob.hanna[at]weizmann.ac.il

Hartig, Leonie
GSI Helmholtzzentrum für Schwerionenforschung GmbH, Darmstadt
l.hartig[at]gsi.de

Hauser, Stefan
German Center for Neurodegenerative Diseases, Tübingen
stefan.hauser[at]dzne.de

Heiduschka, Sonja
University Hospital Düsseldorf, Düsseldorf
sonja.heiduschka[at]med.uni-duesseldorf.de

Heil, Sarah
ISAR Bioscience GmbH, Planegg
sarah.heil[at]isarbioscience.de

Heise, Dirk
Biozym Vertrieb GmbH, Hess. Oldendorf
dirk.heise[at]biozym.com

Hempfling, Detlef
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
detlefh[at]miltenyibiotec.de

Henning, Nina
Leibniz Institute on Aging (FLI), Jena
nina.henning[at]leibniz-fli.de

Hernandez Bautista, Carlos Alberto
Hannover Medical School, Hannover
hernandezbautista.carlos[at]mh-hannover.de

Herrmann, Ira
LIFE & BRAIN GmbH, Bonn
iherrmann[at]lifeandbrain.com

Herrmann, Sabrina
University of Saarland/Fraunhofer IBMT, Würzburg
sabrina.herrmann[at]ibmt.fraunhofer.de

Heuberger, Julian
German Federal Institute for Risk Assessment, Berlin
julian.heuberger[at]bfr.bund.de

Hiller, Björn
Catalent Düsseldorf GmbH, Langenfeld
bjoern.hiller[at]catalent.com

Hinkelmann, Alica
Evotec, Göttingen
alica.hinkelmann[at]evotec.com

Hofmann, Samuel
Center for Child and Adolescent Medicine, University Hospital Heidelberg, Heidelberg
samuel.hofmann[at]med.uni-heidelberg.de

Hohl, Anastassja
Thermo Fisher Scientific, Darmstadt
anastassja.hohl[at]thermofisher.com

Höhne, Karin
Berlin Institut of Health, Berlin
karin.hoehne[at]bih-charite.de

Hohwieler, Meike
Institute of Molecular Oncology and Stem Cell Biology, Ulm
meike.hohwieler[at]uni-ulm.de

Hoja, Matthias
iotaSceinces Ltd, Leipzig
mhoja[at]iotasciences.com

Horland, Reyk
TissUse GmbH, Berlin
reyk.horland[at]tissuse.com

Horlova, Anastasiia
EMBL, Heidelberg
anastasiia.horlova[at]embl.de

Hörmanseder, Eva
Helmholtz Munich, Munich
eva.hoermanseder[at]helmholtz-munich.de

Horschitz, Sandra
Central Institute of Mental Health, Mannheim
sandra.horschitz[at]zi-mannheim.de

Hradil, Vojtech
Leibniz Research Laboratories for Biotechnology and Artificial Organs, Hannover
hradil.vojtech[at]mh-hannover.de

Hrynchak, Mariya
Nippon Geneticse Europe GmbH, Düren
mhrynchak[at]nippongenetics.de

Hufschmid, Katja
Thermo Fisher Scientific, Reinach, CH
katja.hufschmid[at]thermofisher.com

Hurley, Killian
Royal College of Surgeons in Ireland, Dublin, IE
killianhurley[at]rcsi.ie

Hussein, Mohamed
Dubai Medical University, College of Medicine, Dubai, AE
dr.m.hussin[at]dmu.ae

I

Icheln, Bärbel
Active Bioscience, Hamburg
icheln[at]active-bioscience.de

Icheln, Johanna
Active Bioscience GmbH, Hamburg
info[at]active-bioscience.de

Inaas, Syeda
Uniklinik RWTH Aachen, Aachen
sinaas[at]ukaachen.de

Iranpour, Sonia
Osnabrück University, Osnabrück
sonia.iranpour[at]yahoo.com

Ivics, Zoltan
Fraunhofer Institute for Cell Therapy and Immunology, Leipzig
zoltan.ivics[at]izi.fraunhofer.de

J

Jabali, Ammar
ISAR Bioscience, Planegg
ammar.jabali[at]isarbioscience.de

Jacobs, Mirta
Cellseeker, Hamburg
mirta[at]accellerate.me

Jauch, Ralf
School of Biomedical Science, Hong Kong, CN
ralf[at]hku.hk

Jeremias, Irmela
Helmholtz Munich, Munich
irmela.jeremias[at]helmholtz-munich.de

Jerred, Caleb
University Hospital Düsseldorf, Düsseldorf
caljerred[at]outlook.com

Jeruzalska, Estera
Institute of Pharmaceutical Biotechnology, Ulm
estera.pluzek[at]uni-ulm.de

Jiang, Yajuan
Institute of Physiology and Pathophysiology of the Medical Faculty- University of Cologne
Marga and Walter Boll Laboratory for Cardiac Tissue Engineering, Cologne
jiangyajuan01[at]outlook.com

Jin, Qianren
Lund University, Lund, SE
qianren.jin[at]med.lu.se

Johannsen, Hannah
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
hannahj[at]miltenyi.com

Johansson, Pia
Lund University, Lund, SE
pia.johansson[at]med.lu.se

Jolly, Adrien
University Hospital Frankfurt, Frankfurt am Main
a.jolly[at]dkfz.de

Jung-Klawitter, Sabine
Center for Child and Adolescent Medicine, University Hospital Heidelberg, Heidelberg
sabine.jung-klawitter[at]med.uni-heidelberg.de

Jung, Jessica
Central Institute of Mental Health (CIMH), Mannheim
jessica.jung[at]zi-mannheim.de

Jung, Johannes
Sartorius Stedim Biotech, Hattersheim
johannes.jung[at]sartorius.com

Jung, Matthias
Institute of Physiological Chemistry (IPC), Halle (Saale)
matthias.jung[at]uk-halle.de

Jungwirth, Linda
Proteintech, Planegg-Martinsried
linda.jungwirth[at]ptglab.com

K

Kaesz, Philip
Biomerieux, Nürtingen
philip.kaesz[at]biomerieux.com

Kafadar, Selen
TissUse GmbH, Berlin
selen.kafadar[at]tissuse.com

Kalayar Aye, Saw
Chiang Mai University, Thailand, Chiang Mai, TH
sawkalayaraye_saw[at]cmu.ac.th

Kamal, Aryan
University of Basel, Basel, CH
aryan.kamal[at]unibas.ch

Karampelias, Christos
Helmholtz Center Munich, Oberschleissheim
christos.karampelias[at]helmholtz-munich.de

Karnatz, Isabell
Saarland University, Würzburg
isabell.karnatz[at]ibmt.fraunhofer.de

Kaysan, Leon
GSI Helmholtzzentrum für Schwerionenforschung GmbH, Darmstadt
l.kaysan[at]gsi.de

Kelle Özdemir, Dolunay
Helmholtz Munich, Munich
dolunay.kelleoezdemir[at]helmholtz-munich.de

Keßler, Antonia
LMU, Faculty of Biology, Munich
a.kessler[at]bio.lmu.de

Ketteler, Carolin
Institute for Structural Biology, Neuherberg
carolin.ketteler[at]helmholtz-munich.de

Khan, Sohana Islam
Uniklinikum Halle, Halle (Saale)
sohana.khan[at]uk-halle.de

Khmelnytska, Yuliia
LLC Medical Center, Reocell, Kyiv, UA
yuliia.khmelnytska[at]reocell.com

Kim, Johnny
TRON, Mainz
johnny.kim[at]tron-mainz.de

Kim, Kee-Pyo
The Catholic University of Korea, Seoul, KR
kpkim[at]catholic.ac.kr

Kipper, Sophie
Helmholtz Munich, Neuherberg
sophie.kipper[at]helmholtz-munich.de

Klebl, Bert
Lead Discovery Center GmbH, Dortmund
klebl[at]lead-discovery.de

Kleemiß, Felix
LEBAO, Hannover
kleemiss.felix[at]mh-hannover.de

Kleger, Alexander
Ulm University, Ulm
alexander.kleger[at]uni-ulm.de

Klein, Dominik
Institute of Computational Biology, Munich
dominik.klein[at]helmholtz-munich.de

Klump, Hannes Joachim
University Hospital Aachen, Aachen
hklump[at]ukaachen.de

Koch, Philipp
Hector Institute for Translational Brain Research, Mannheim
philipp.koch[at]zi-mannheim.de

Köhl, Ulrike
Fraunhofer Institute for Cell Therapy and Immunology, Leipzig
ulrike.koehl[at]izi.fraunhofer.de

Kollenda-Strunk, Sebastian
SYNENTEC GmbH, Elmshorn
s.kollenda-strunk[at]synentec.com

Kopplin, Robert
Osnabrück University, Osnabrück
robert.kopplin[at]uni-osnabrueck.de

Koufali, Angeliki
Pharmacology and Toxicology, Göttingen
angeliki.koufali[at]med.uni-goettingen.de

Kranz, Andrea
Biotec, Dresden
andrea.kranz[at]tu-dresden.de

Krietemeyer, Björn
Nippon Genetics Europe GmbH, Düren
bkrietemeyer[at]nippongenetics.de

Krija, Melissa
Leibniz Institute on Aging Research, Jena
melissa.krija[at]leibniz-fli.de

Kroeger, Joseph
ISAR Bioscience, Planegg
joseph.kroeger[at]isarbioscience.de

Krümpelmann, Laura
Roche Diagnostics GmbH, Penzberg
laura.kruempelmann[at]roche.com

Krutenko, Tamara
Institute of Reconstructive Neurobiology, Bonn
krutenko[at]uni-bonn.de

Kuleshova, Anna
Hanover Medical School, Hannover
kuleshova.anna[at]mh-hannover.de

Külp, Marius
Goethe University Hospital Frankfurt, Frankfurt am Main
marius.kuelp[at]em.uni-frankfurt.de

Kuninger, David
Thermo Fisher Scientific, Frederick, US
david.kuninger[at]thermofisher.com

Künne, Andreas
DLR Projektträger, Cologne
andreas.kuenne[at]dlr.de

Kurtz, Andreas
BIH Center for Regenerative Therapies, Berlin
andreas.kurtz[at]charite.de

Kurz, Konstantin
BioThrust GmbH, Aachen
konstantin.kurz[at]biothrust.de

Kuzmich, Sofya
Sartorius Lab Instruments GmbH & Co. KG, Göttingen
sofya.kuzmich[at]sartorius.com

L

Lachmann, Nico
Hannover Medical School (MHH), Hannover
lachmann.nico[at]mh-hannover.de

Lang, Sarah
Hi-STEM gGmbH, Heidelberg
sarah.lang[at]dkfz-heidelberg.de

Ladewig, Julia
Central Institute of Mental Health (CIMH), Mannheim
julia.ladewig[at]zi-mannheim.de

Langer, Astrid
European Patent Office, Munich
alanger[at]epo.org

Laufer, Sandra
University Hospital Hamburg-Eppendorf, Hamburg
s.laufer[at]uke.de

Laugwitz, Karl-Ludwig
Klinikum rechts der Isar, TU Munich, Munich
laugwitz[at]mytum.de

Lemme, Marta
Nanion Technologies, Munich
marta.lemme[at]nanion.de

Li, Rui
TRON (Translational Oncology), Mainz
rui.li[at]tron-mainz.de

Liang, Manqi
Ludwig Maximilian University Munich, Munich
dayin.dayin111[at]gmail.com

Lickert, Heiko
Helmholtz Munich, Neuherberg
heiko.lickert[at]helmholtz-muenchen.de

Lim, Bryce
German Cancer Research Center (DKFZ), London, UK
ecyrblim[at]gmail.com

Lim, Sunny
OG Hospital, Medan, ID
dr.sunnylim74[at]yahoo.com

Lisboa, Ana
Thermo Fisher Scientific, Madrid, ES
ana.lisboa[at]thermofisher.com

Lithovius, Väinö
Helmholtz Munich, Munich
vaino.lithovius[at]helmholtz-munich.de

Loser, Michael
Ibidi, Gräfelfing
mloser[at]ibidi.de

Loss, Isabel
Heidelberg University, Mannheim
isabel.loss[at]medma.uni-heidelberg.de

Lu, Tianlin
TRON gGmbH, Mainz
tianlin.lu[at]tron-mainz.de

Lu, Ziwei
University of Cologne, Medical Faculty, Cologne, Germany, Cologne
lu_ziwei[at]yeah.net

Lubitz, Sandra
Evotec, Hamburg
sandra.lubitz[at]evotec.com

Luger, Elke
National Network Office GCT Germany, Berlin
elke.luger[at]bih-charite.de

Luo, Yanyan
Charité –Berlin University Medicine, Berlin
yanyan.luo[at]charite.de

Luong, Ngoc
Central Institute of Mental Health, Mannheim
ngoc.luong[at]zi-mannheim.de

M

Mach, Benedikt
ISAR Bioscience GmbH, Planegg
benedikt.mach[at]isarbioscience.de

Mahler, Stefanie
German Stem Cell Network (GSCN), Berlin
stefanie.mahler[at]bih-charite.de

Maier, Alexander
Miltenyi Biotec B.V. & Co. KG, Bergisch Gladbach
alexandermai[at]miltenyibiotec.de

Maiwald, Julia Luisa
Pharmacology & Toxicology, University Hospital Göttingen, Göttingen
maiwaldjulia13[at]gmail.com

Mall, Moritz
German Cancer Research Center (DKFZ), Heidelberg
m.mall[at]dkfz.de

Mansour, Gizem
Helmholtz Pioneer Campus, Oberschleißheim
gizem.mansour[at]helmholtz-munich.de

Marcato, Silvia
Helmholtz Munich, Munich
silvia.marcato[at]helmholtz-munich.de

Mark, Peter
Thermo Fisher Scientific, Darmstadt
peter.mark[at]thermofisher.com

Markova, Pavlina
EMBL, Heidelberg
pavlina.markova[at]embl.de

Marr, Carsten
Helmholtz Munich, Neuherberg
carsten.marr[at]helmholtz-muenchen.de

Martin-Villalba, Ana
German Cancer Research Center (DKFZ), Heidelberg
a.martin-villalba[at]dkfz.de

Martin, Ulrich
Hannover Medical School (MHH), Hannover
martin.ulrich[at]mh-hannover.de

Martins, Inês
Max Delbrück Center for Molecular Medicine, Berlin
ines.afonsomartins[at]mdc-berlin.de

Martins, Soraia
Catalent Düsseldorf GmbH, Düsseldorf
soraia.martins[at]catalent.com

Mass, Elvira
University of Bonn, Bonn
emass[at]uni-bonn.de

Mastalerz, Michal
Novogene Europe, Planegg
michal.mastalerz[at]novogene-europe.com

Matros, Julia
FUJIFILM Wako Chemicals, Neuss
julia.matros[at]fujifilm.com

Maurer, Jochen
University Hospital RWTH Aachen, Aachen
jmaurer[at]ukaachen.de

Meier, Magdalena
PELOBiotech GmbH, Planegg
magdalena.meier[at]pelobiotech.com

Méndez Muñoz, Carla
Helmholtz Munich, Munich
carla.mendez-munoz[at]helmholtz-munich.de

Merkert, Sylvia
Hannover Medical School (MHH), Hannover
merkert.sylvia[at]mh-hannover.de

Metz, Isabell
BIOZOL Diagnostica Vertrieb GmbH, Hamburg
imetz[at]biozol.de

Metzger, Hugo
MHH, Hannover
metzger.hugo[at]mh-hannover.de

Metzger, Jakob
Max Delbrück Center (MDC), Berlin
jakob.metzger[at]mdc-berlin.de

Meurer, Marita
University of Veterinary Medicine, Institute of Biochemistry, Hannover
marita.meurer[at]tiho-hannover.de

Meyer, Thorben
Active Bioscience GmbH, Hamburg
info[at]active-bioscience.de

Milsom, Michael
HI-STEM gGmbH, Heidelberg
m.milsom[at]hi-stem.de

Minaeva, Mariia
Hemholtz Munich, Munich
mariia.minaeva[at]helmholtz-munich.de

Miranda, Martin
Helmholtz, Munich
mrtn.mirnd[at]gmail.com

Molho, Chiara
ETH Zürich, Zurich, CH
cmolho[at]ethz.ch

Molina Van den Bosch, Mireia
Helmholtz Munich, Munich
mireia.molina[at]helmholtz-munich.de

Mönkemöller, Raphael
Institute for Mechatronic Systems, Garbsen
raphael.moenkemoeller[at]imes.uni-hannover.de

Moretti, Alessandra
TUM School of Medicine and Health, Munich
amoretti[at]mytum.de

Muhammad, Aisha Umar
Institute of Translational Stem Cell, Munich
aisha.muhammad[at]helmholtz-munich.de

Münchhalfen, Lisa
Marga& Walter Boll Laboratory for Cardiac Tissue Engineering, Cologne
lisamuenchhalfen[at]gmail.com

Mytilineos, Joannis
German Central Stem Cell Donor Registry (ZKRD), Ulm
joannis.mytilineos[at]zkrd.de

N

Nagy, Andras
Sinaí Health System, Toronto, CA
tapia[at]lunenfeld.ca

Natrup, Nils
Hannover Medical School (MHH), Hannover
Nils[at]natrup.de

Naujock, Maximilian
Evotec, Goettingen
maximilian.naujock[at]evotec.com

Nawroth, Janna
Helmholtz Pioneer Campus, Neuherberg
janna.nawroth[at]helmholtz-munich.de

Nebol, Aylin
Max Delbrück Center, Berlin
aylin.nebol[at]mdc-berlin.de

Nenninger, Beate-Sophie
Saarland University, Würzburg
beate-sophie.nenninger[at]ibmt.fraunhofer.de

Neubauer, Julia
Fraunhofer Institute for Biomedical Engineering IBMT, Sulzbach
julia.neubauer[at]ibmt.fraunhofer.de

Neumann, Katrin
Center for Regenerative Therapies Dresden (CRTD), Dresden
katrin.neumann1[at]tu-dresden.de

Niemann, Julian
Institute for Molecular Medicine, Ulm
julian.niemann[at]uni-ulm.de

Niemöller, Theresa
Leibniz Research Laboratories for Biotechnology and Artificial Organs, Hannover
niemoeller.theresa[at]mh-hannover.de

Niessing, Dierk
Institute of Pharmaceutical Biotechnology, Ulm
dierk.niessing[at]uni-ulm.de

Nischalke, Vera
Institute of Stem Cell Biology, Aachen
vnischalke[at]ukaachen.de

Nold, Philipp
Eppendorf, Juelich
nold.p[at]eppendorf.de

Nowak, Katja
University of Bielefeld, Bielefeld
katja.nowak[at]uni-bielefeld.de

Nugraha, Osman Abhimata
Institute for Medical Informatics and Biometry (IMB), Dresden
osman_abhimata.nugraha[at]tu-dresden.de

Nurieva, Wasifa
UKSH Campus Lübeck, Childrens Hospital, Lübeck
wasifa.nurieva[at]uksh.de

Nürnberg, Elina
faCellitate GmbH, Mannheim
elina.nuernberg[at]facellitate.com

O

Obser, Robin
Eppendorf, Juelich
obser.r[at]eppendorf.de

Özata, Gamze
LMU-DZNE, Munich
gamze.oezata[at]mail03.med.uni-muenchen.de

Olmer, Ruth
Hannover Medical School (MHH), Hannover
olmer.ruth[at]mh-hannover.de

Olmos, Julian
PELOBiotech GmbH, Planegg
julian.olmos[at]pelobiotech.com

Orschmann, Tanja
Core Facilities, Munich
tanja.orschmann[at]helmholtz-munich.de

Ozaki, Mari
Royal College of Surgeons in Ireland, Dublin, IE
mariozaki[at]rcsi.ie

P

Panza, Paolo
Justus Liebig University Giessen, Bad Nauheim
paolo.panza[at]uni-giessen.de

Paquet, Dominik
University Hospital, LMU Munich, Munich
dominik.paquet[at]med.uni-muenchen.de

Pérez Fernández, Raquel
Central Institute of Mental Health, Mannheim
raquel.perezfernandez[at]zi-mannheim.de

Perez-Munoz, Sara
Thermo Fisher Scientific, Madrid, ES
sara.perez[at]thermofisher.com

Persson, Elin
BioLamina AB, Sundbyberg, SE
elin.persson[at]biolamina.com

Petzold, Heike
MPI-CBG, Dresden
hpetzold[at]mpi-cbg.de

Pezzica, Tommaso
Evotec International GmbH, Göttingen
tommaso.pezzica[at]evotec.com

Pfaender, Stefanie
Evotec, Göttingen
stefanie.pfaender[at]evotec.com

Pfannkuche, Kurt
Zentrum für Physiologie, Cologne
kurt.pfannkuche[at]uni-koeln.de

Pfeiffer, Matthias
Sarstedt Deutschland, Nümbrecht
matthias.pfeiffer[at]sarstedt.com

Pförtner, Felix
LMU, Faculty of Biology, Neuried
pfoertner[at]bio.lmu.de

Piazzalonga, Francesca
IMOS, Ulm
francesca.piazzalonga[at]uni-ulm.de

Piraud, Marie
Helmholtz Munich, Oberschleißheim
marie.piraud[at]helmholtz-munich.de

Plesnik, Sara
LMU Faculty for Biology, Planegg
plesnik[at]bio.lmu.de

Pless, Ole
Fraunhofer Institute for Translational Medicine and Pharmacology ITMP, Hamburg
ole.pless[at]itmp.fraunhofer.de

Plötner, Clara Luise
Hannover Medical School (MHH), Hannover
ploetner.clara[at]mh-hannover.de

Podlesnic, Martina
Molecular Biology, Innsbruck, AT
martina.podlesnic[at]uibk.ac.at

Pohl, Corinna
Thermo Fisher Scientific, Darmstadt
corinna.pohl[at]thermofisher.com

Puertollano, David
Hannover Medical School (MHH), Hannover
puertollanomartin.david[at]mh-hannover.de

Puri, Deepika
Institute for Stem Cell Biology, Uniklinik, Aachen, Aachen
dpuri[at]ukaachen.de

Q

Quach, Susanna
Charite, Berlin
susanna.quach[at]charite.de

R

Rädecke, Kristin
Institute of Pharmacology, Heidelberg
kristin.raedecke[at]pharma.uni-heidelberg.de

Rafiei Hashtchin, Anna
Max-Delbrück-Center for Molecular Medicine, Berlin
anna.rafieihashtchin[at]mdc-berlin.de

Rebola Lameira, Maria Leonor
TRON gGmbH, Mainz
leonor.lameira[at]tron-mainz.de

Reigl, Amelie
Fraunhofer ISC TLZ-RT, Würzburg
amelie.reigl[at]isc.fraunhofer.de

Reinelt, Anna
University Medical Center Hamburg-Eppendorf, Hamburg
a.reinelt[at]uke.de

Reinhardt, Peter
AbbVie Deutschland GmbH & Co KG, Ludwigshafen
peter.reinhardt[at]abbvie.com

Restle, Anja
Cenibra GmbH, Bramsche
anja.restle[at]cenibra.de

Richter, Lisa
Becton Dickinson, Heidelberg
lisa.richter[at]bd.com

Richter, Sven
TUM German Heart Center Munich, Munich
sven.richter3[at]gmx.de

Rieger, Michael
University Hospital Frankfurt, Frankfurt/Main
m.rieger[at]em.uni-frankfurt.de

Risse, Isabelle
Institute of Physiology I, Bonn
iris[at]uni-bonn.de

Rivandi, Mahdi
MedChemExpress, Sollentuna, SE
mahdi.rivandi[at]medchemexpress.com

Rodrigues Pereira, Osvaldo
Institute of Diabetes and Regeneration, Munich
osvaldo.pereira[at]helmholtz-munich.de

Roeder, Ingo
TU Dresden, Dresden
ingo.roeder[at]tu-dresden.de

Rogers, Nicole
Helmholtz Diabetes Center, Neuherberg
nicole.rogers[at]helmholtz-munich.de

Romeike, Merrit
Berlin Institute of Health at Charité, Berlin
merrit.romeike[at]bih-charite.de

Romero-Mulero, Mari Carmen
ETH Zürich, Zurich, CH
mariadelcarmen.romeromulero[at]hest.ethz.ch

Roth, Steffen
Xceltis GmbH, Mannheim
info[at]xceltis.de

Rusha, Ejona
Helmholtz Munich, Pfaffenhofen
ejona.rusha[at]helmholtz-munich.de

S

Sabihi Ahvaz, Zohre
Hannover Medical School, Hannover
sabihiahvaz.zohre[at]mh-hannover.de

Sydler, Becky
VectorBuilder, Neu-Isenburg
beckysadler[at]vectorbuilder.com

Saleh, Fawaz
Department of Pediatric Pneumology, Allergology and Neonatology, Hannover
saleh.fawaz[at]mh-hannover.de

Sardogan, Aydan
Institute of Lung Health and Immunity, Neuherberg
aydan.sardogan[at]helmholtz-munich.de

Saric, Tomo
Center for Physiology and Pathophysiology, Cologne
tomo.saric[at]uni-koeln.de

Scamozzi, Maria
Institute of Translational Stem Cell, Munchen
maria.scamozzi[at]helmholtz-munich.de

Scarcella, Daniele
Helmholtz Munich, Munich
daniele.scarcella[at]helmholtz-munich.de

Schaller, Heidrun
Forschungslabor Hämatologie und Zelltherapie, Leipzig
heidrun.schaller[at]gmx.de

Scheinpflug, Philine
Evotec, Göttingen
philine.scheinpflug[at]evotec.com

Schlegel, Timm
Axion BioSystems, Eindhoven,NL
tschlegel[at]axionbio.com

Schlichthärle, Thomas
TU Munich, Garching
thomas.schlichthaerle[at]tum.de

Schmachtel, Tessa
Goethe University Frankfurt, Frankfurt am Main
schmachtel[at]med.uni-frankfurt.de

Schmelz, Karin
Charité, Berlin
karin.schmelz[at]charite.de

Schmidt, Aileen
Medical school hannover (MHH), Hannover
schmidt.aileen[at]mh-hannover.de

Schmidt, Malin
Zentralinstitut für Seelische Gesundheit, Mannheim
malin.schmidt[at]zi-mannheim.de

Schmoll, Kea
University-Medical Center Goettingen, Goettingen
keaaline.schmoll[at]med.uni-goettingen.de

Schöler, Hans
Max-Planck-Institut für molekulare Biomedizin, Münster
h.schoeler[at]mpi-muenster.mpg.de

Schröder, Insa
GSI Helmholtzzentrum für Schwerionenforschung, Darmstadt
i.schroeder[at]gsi.de

Schroeder, Timm
ETH Zurich, Basel (BSSE), Basel
timm.schroeder[at]bsse.ethz.ch

Schröter, Christian
Leiden Academic Centre for Drug Research, Oegstgeest, NL
c.schroter[at]lacdr.leidenuniv.nl

Schultz, Ursula
Sartorius CellGenix GmbH, Freiburg
ursula.schultz[at]sartorius.com

Schulz, Gert
Bio-Techne, Wiesbaden
gert.schulz[at]bio-techne.com

Schumann, Gerald G.
Paul Ehrlich Institute, Langen
gerald.schumann[at]pei.de

Schupp, Jonas
Hannover Medical School, Hannover
schupp.jonas[at]mh-hannover.de

Schurer, Amelie
Universität Innsbruck, Götzens, AT
amelie.schurer[at]uibk.ac.at

Schüring, Max-Peter
Sartorius Lab Instruments GmbH & Co. KG, Göttingen
max-peter.schuering[at]sartorius.com

Schuster, Felix
ISAR Bioscience GmbH, Planegg
felix.schuster[at]isarbiocience.de

Schwarz-Romond, Thomas
Helmholtz Pioneer Campus, Neuherberg
thomas.schwarz-romond[at]helmholtz-munich.de

Scialdone, Antonio
Helmholtz Munich, Munich
antonio.scialdone[at]helmholtz-munich.de

Segeletz, Sandra
faCellitate GmbH, Mannheim
sandra.segeletz[at]facellitate.com

Seltmann, Stefanie
Universitätsklinikum Heidelberg, Heidelberg
stefanie.seltmann[at]med.uni-heidelberg.de

Shaheen, Sameerah
King Saud University, Riyadh, SA
sshahee[at]ksu.edu.sa

Shamshur, Maryna
LLC Medical Center, Reocell, Kyiv, Ukraine
maryna.shamshur[at]gmail.com

Shelashen, Arina
TU Dresden, Dresden
arina.shelashen[at]tu-dresden.de

Shigematsu, Kazuki
PeptiGrowth Inc., Tokyo, JP
kazuki.shigematsu[at]peptigrowth.com

Silva, Miguel
TUM, Munich
miguel.silva[at]tum.de

Singh, Niraj Kumari
Deutsches Herzzentrum Munich, Hemsbach
nir.raj12singh[at]gmail.com

Sluijter, Joost
University Medical Center Utrecht, Utrecht, NL
j.sluijter[at]umcutrecht.nl

Sommer, Annika
Roche Diagnostics, Penzberg
annika.sommer[at]roche.com

Spathopoulou, Angeliki
Molecular biology, Innsbruck, AT
angeliki.spathopoulou[at]uibk.ac.at

Speiseder, Thomas
Thermo Fisher Scientific, Darmstadt
thomas.speiseder[at]thermofisher.com

Srinidhi, Jahnavi
University clinic Mannheim, Heidelberg University, Mannheim
jahnavi.srinidhi[at]medma.uni-heidelberg.de

Stachelscheid, Harald
Charité - Universitätsmedizin Berlin, Berlin
harald.stachelscheid[at]charite.de

Staubach, Simon
Sartorius, Cologne
simon.staubach[at]sartorius.com

Steeb, Lothar
PELOBiotech GmbH, Planegg
lothar.steeb[at]pelobiotech.com

Steenpaß, Laura
Leibniz-Institut DSMZ, Braunschweig
laura.steenpass[at]dsmz.de

Steinheimer-Breitkreutz, Konstanze
European Patent Office, Munich
ksteinheimerbreitkreutz[at]epo.org

Sterneckert, Jared
Technische Universität Dresden, Dresden
jared.sterneckert[at]tu-dresden.de

Sterr, Michael
Helmholtz Munich, Neuherberg
michael.sterr[at]helmholtz-munich.de

Stewart, Liz
Qkine, Cambridge, UK
liz.stewart[at]qkine.com

Stock, Marco
Helmholtz Zentrum Munich, Neuherberg
marco.stock[at]helmholtz-munich.de

Straßner, Elena
CMCB, BIOTEC, Dresden
elena.strassner[at]tu-dresden.de

Stricker, Stefan
LMU Munich, Munich
stefan.stricker[at]helmholtz-muenchen.de

Strunk, Dirk
Experimental and Clinical Cell Therapy, Salzburg, AT
cornelia.adhero[at]pmu.ac.at

Stüdemann, Tim
TRON gGmbH, Mainz
tim.stuedemann[at]tron-mainz.de

Sukhoverkhov, Arkhyp
ISAR Bioscience, Munich
ammar.jabali[at]gmx.de

T

Tabar, Viviane
Memorial Sloan Kettering Cancer Center, New York
tabarv[at]mskcc.org

Taxauer, Karin
BIOZOL Diagnostica Vertrieb GmbH, Hamburg
karin.taxauer[at]biozol.de

Teichert, Ferdinand
Medizinsiche Hochschule Hannover & Leiden University, Pullach
ferdlteichert[at]googlemail.com

Terheyden-Keighley, Daniel
Catalent, Langenfeld
daniel.keighley[at]catalent.com

Tew, Hui Ming
Goethe University Hospital Frankfurt, Frankfurt am Main
tew[at]med.uni-frankfurt.de

Thalhammer, Stefan
ISAR Bioscience GmbH, Planegg
stefan.thalhammer[at]isarbioscience.de

Thomanetz, Venus
Abbvie GmbH&Ko.KG, Ludwigshafen
venus.thomanetz[at]abbvie.com

Tian, Chenglei
Institute of Translational Stem Cell Research, Neuherberg
cltian0401[at]gmail.com

Tirandazi, Vista
Osnabrück University, Osnabrück
vtirandazi[at]uni-osnabrueck.de

Tjoeng, Iva
Evotec SE, Hamburg
iva.tjoeng[at]evotec.com

Tolle, Isabella
Medical Faculty, Heinrich Heine University Düsseldorf, Düsseldorf
isabella.tolle[at]med.uni-duesseldorf.de

Tonn, Torsten
DRK-Blutspendedienst Baden-Württemberg - Hessen gGmbH, Frankfurt/Main
t.tonn[at]blutspende.de

Torres-Padilla, Maria Elena
Helmholtz Zentrum Munich, Neuherberg
torres-padilla[at]helmholtz-muenchen.de

Trillhaase, Anja
Breitenfelde
anja-trillhaase[at]gmx.de

Trpchevska, Ana
Fraunhofer ITMP, Göttingen
anatrpchevska15[at]outlook.com

Trumpp, Andreas
HI-STEM gGmbH, Heidelberg
a.trumpp[at]dkfz-heidelberg.de

U

Uhlin, Elias
BioLamina, Sundbyberg, SE
elias.uhlin[at]biolamina.com

Uhrig, Markus
OMNI Life Science GmbH & Co. KG, Bremen
markus.uhrig[at]ols-bio.de

Ureche, Cristina I.
Minaris Advanced Therapies, Ottobrunn
cristina.ureche[at]advancedtherapies.com

Uxa, Sigrid
University Leipzig, Leipzig
sigrid.uxa[at]medizin.uni-leipzig.de

V

Velazco Stock, Nicole
Merck KGaA, Hamburg
nicolevelazco[at]gmail.com

Veldhues, Antje
German Stem Cell Network (GSCN), Berlin
antje.veldhues[at]bih-charite.de

Vincendeau, Michelle
Helmholtz Munich, Oberschleißheim
michelle.vincendeau[at]helmholtz-munich.de

W

Wagner, Wolfgang
Rheinisch-Westfälische Technische Hochschule Aachen, Aachen
wwagner[at]ukaachen.de

Wang, Danni
Novogene Europe, Planegg
danni.wang[at]novogene-europe.com

Waskow, Claudia
Leibniz Institute on Aging, Jena
claudia.waskow[at]uni-jena.de

Wasnyck, Roxana
Institute of Lung Health and Immunity, Untermeitingen
roxanamaria.wasnick[at]helmholtz-munich.de

Wehmeier, Oliver
Cellseeker, Hamburg
oliver[at]accellerate.me

Weller, Arjen
Fraunhofer ITMP, Hamburg
arjen.weller[at]itmp.fraunhofer.de

Wessel, Tim
Thermo Fisher Scientific, Darmstadt
tim.wessel[at]thermofisher.com

Wilbertz, Johannes
Ksilink Services SAS, Strasbourg, FR
johannes.wilbertz[at]ksilink.com

Wilkinson, Adam
University of Cambridge, Cambridge, UK
acw63[at]cam.ac.uk

Wilson, Liam
Hannover Medical School, Hannover
wilson.liam[at]mh-hannover.de

Witt, Skyla
ISAR Bioscience, Planegg
skyla.witt[at]isarbioscience.de

Wittich, Annika
Fraunhofer ITMP, Hamburg
annika.wittich[at]itmp.fraunhofer.de

Wörner, Nils
HI-STEM gGmbH, Heidelberg
nils.woerner[at]dkfz-heidelberg.de

X

Xu, Wenqing
Heinrich Heine University düsseldorf, Düsseldorf
xuwenqingxwq[at]gmail.com

Xu, Yanjun
Helmholtz Center Munich, Munich
yanjun.xu[at]helmholtz-munich.de

Xue, Meiying
Charité – Universitätsmedizin Berlin, Berlin
meiying.xue[at]charite.de

Y

Yaman, Ferhat Ali
Berlin Institute of Health at Charité, Berlin
ferhat-ali.yaman[at]charite.de

Yan, Yaobin
TRON gGmbH, Mainz
yaobin.yan[at]tron-mainz.de

Z

Zarvandi, Samira
Human Genetics, Hannover
zarvandi.samira[at]mh-hannover.de

Zeevaert, Kira
Institute of Stem Cell Biology, Aachen
kzeevaert[at]ukaachen.de

Zenagui, Reda
Stem Genomics, Grabels, FR
reda.zenagui[at]stemgenomics.com

Zenke, Martin
Department of Medicine IV, Aachen
martin.zenke[at]rwth-aachen.de

Zerhusen, Christian
Eppendorf, Juelich
zerhusen.c[at]eppendorf.de

Zerjatke, Thomas
TU Dresden, Dresden
thomas.zerjatke[at]tu-dresden.de

Zimmer, Bastian
Evotec SE, Hamburg
bastian.zimmer[at]evotec.com

Zimmermann, Pauline
DLR Project Management Agency, Berlin
pauline.zimmermann[at]dlr.de

Zimmermann, Wolfram-Hubertus
University Medical Center Göttingen, Göttingen
w.zimmermann[at]med.uni-goettingen.de

Zink, Annika
Universitätsklinikum Düsseldorf, Düsseldorf
annika.zink[at]med.uni-duesseldorf.de

Zweigerdt, Robert
Medizinische Hochschule Hannover, Hannover
zweigerdt.robert[at]mh-hannover.de

Zywitza, Vera
Leibniz-Institut for Zoo and Wildlife Research
(Leibniz-IZW) in the Forschungsverbund Berlin
zywitza[at]izw-berlin.de

Supporter & exhibitors

Academic Supporters

Helmholtz Munich
Ingolstädter Landstraße 1
85764 Neuherberg
www.helmholtz-munich.de/en/

German Research Foundation (DFG)
Kennedyallee 40
53175 Bonn
www.dfg.de/en

Berlin Institute of Health (BIH)
@ Charité – Universitätsmedizin Berlin
Anna-Louisa-Karsch-Straße 2
10178 Berlin
www.bihealth.org

National Joint Project TACTiC
Hannover Medical School (MHH), OE 6876
Carl-Neuberg-Straße 1
30625 Hannover
www.mhh.de/en/lebao/research/tactic

DZL Research Network BREATH
Member of the German Center for Lung Research (DZL)
Hannover Medical School (MHH), OE 6876
Carl-Neuberg-Straße 1
30625 Hannover
www.breath-hannover.de/en/nc.htm

Gold Supporters



STEMCELL Technologies GmbH
Stolberger Str. 200
50933 Cologne
www.stemcell.com

Silver Supporters



Active Bioscience GmbH
Oberaltenallee 8
D-22081 Hamburg
www.active-bioscience.de



Eppendorf AG
Bioprocess Center
Rudolf-Schulten-Str. 5
52428 Jülich
www.eppendorf.com



FUJIFILM Wako Chemicals Europe GmbH
Fuggerstraße 12
41468 Neuss
www. wako-chemicals.de



PeptiGrowth Inc.
Nihonbashi Life Science Building 7, 9-10
Nihonbashi Horidomecho 1-chome, Chuo-ku
Tokyo 103-0012
Japan
peptigrowth.com/en/

Bronze Supporters

BioLamina AB
Löfströms Allé 5a
172 66 Sundbyberg
Sweden
www.biolamina.com

bioMérieux Germany GmbH
Weberstraße 8
72622 Nürtingen
www.biomerieux.com

bio-techne
19 Barton Lane
Abingdon | OX1H 3NB
United Kingdom
www.bio-techne.com

Evotec SE
Manfred Eigen Campus
Essener Bogen 7
22419 Hamburg
www.evotec.com

Integrated DNA Technologies (IDT) Germany GmbH
Terminal Straße Mitte 18
85356 München-Flughafen
eu.idtdna.com

iota Sciences
Begbroke Science Park
OX5 1PF Oxford
United Kingdom
www.iotasciences.com

Miltenyi Biotec B.V. & Co. KG
Friedrich-Ebert-Straße 68
51429 Bergisch Gladbach
www.miltenyibiotec.com

Minaris Regenerative Medicine GmbH
Haidgraben 5
85521 Ottobrunn
www.rm.minaris.com

OLS OMNI Life Science GmbH & Co. KG
Karl-Ferdinand-Braun-Straße 2
28359 Bremen
www.ols-bio.de

Qkine
Unit 1, Murdoch House
Garlic Road
Cambridge CB5 8HW
United Kingdom
qkine.com

Sartorius Stedim Biotech GmbH
August-Spindler-Straße 11
37079 Göttingen
www.sartorius.com

stemgenomics
Cap Sigma
1682 rue de la Valsière
4 790 Grabels
France
www.stemgenomics.com

Thermo Fisher Scientific
(formerly Life Technologies)
Frankfurter Straße 129B
64293 Darmstadt
www.thermofisher.com

Exhibitors

acCELLerate GmbH
OsterfelderStr. 12-14
22529 Hamburg
www.accellerate.me

AMS Biotechnology (Europe) Limited
184 Milton Park
Abdington OX144SE
United Kingdom
www.amsbio.com

Becton Dickinson GmbH
BD Biosciences
Tullastraße 8-12
69126 Heidelberg
www.bd.com

Biomol GmbH
Kieler Straße 303a
22525 Hamburg
www.biomol.com

Biozym Scientific GmbH
Steinbrinksweg 27
31840 Hessisch Oldendorf
www.biozym.com

CENiBRA GmbH
Münsterstr. 2
49656 Bramsche
www.cenibra.de

faCellitate GmbH
Franz-Volhard-Str. 5
68167 Mannheim
facellitate.com

ibidi GmbH
Lochhamer Schlag 11
82166 Gräfelfing
ibidi.com

Macherey-Nagel GmbH & Co. KG
Valencienner Straße 11
52355 Düren
www.mn-net.com

MedChemtronica AB
Bergkällvägen 37C
19279 Sollentuna
Sweden
www.medchemexpress.com

New England BioLabs GmbH
Brüningstr. 50, Building B852
65926 Frankfurt am Main
www.neb-online.de

NIPPON Genetics Europe GmbH
Mariaweilerstraße 28 a
52349 Düren
www.nippongenetics.eu

Novogene GmbH
Am Klopferspitz 19a
82152 Planegg
www.novogene.com

PELOBiotech GmbH
Behringstrasse 10
82152 Planegg
www.pelobiotech.com

Proteintech Germany
Fraunhoferstr. 1
82152 Planegg-Martinsried
Germany
www.ptglab.com



SARSTEDT AG & Co. KG
Sarstedtstr. 1
51588 Nümbrecht
www.sarstedt.com



Synentec GmbH
Otto-Hahn-Str. 9a
25337 Elmshorn
synentech.com



TELIGHT Brno, s.r.o.
Škrobárenská 485/14
Building B7 - Unit 3
617 00 Brno
Czech Republic
telight.eu



VectorBuilder GmbH
Hermannstr. 54-56
63263 Neu-Isenburg
en.vectorbuilder.com



XCeltis
Kamenzer Straße 12
68309 Mannheim
www.xceltis.com



ZYMO RESEARCH Europe GmbH
Mülhauser Str. 9
79110 Freiburg
www.zymoresearch.de

Notes

Notes

Notes