SWISS PHARMA SCIENCE DAY 2024

SAPhS
Swiss Academy of Pharmaceutical Sciences

Wednesday 21 August 2024
Von Roll Campus University of Bern

«Challenges and Opportunities in Pharmaceutical Sciences»

Intention

The SWISS PHARMA SCIENCE DAY (SPhSD) is an annual event of the Swiss Academy of Pharmaceutical Sciences (SAPhS, www.saphw.ch). It offers a unique platform for scientists in the field of pharmaceutical sciences to meet, interact and learn.

The 17th SPhSD is placed under the theme of “Challenges and Opportunities in Pharmaceutical Sciences”. Four distinguished invited speakers will address this theme from the perspective of their respective disciplines. As in previous years, MSc and PhD students, as well as Post-Docs of the Swiss Academic Institutions for Pharmaceutical Sciences will have the opportunity to present their latest research in a poster session. Additionally, three poster abstracts will be selected by the scientific committee for a short lecture. A roundtable discussion will close the scientific part, followed by the award ceremony and the traditional apero.

One of the primary goals of the SPhSD is to promote professional and social contacts between students, postdocs and established scientists in industry, academia, hospitals, public health administration, and public pharmacies. For students and young scientists the SPhSD offers a unique platform for learning about opportunities and career paths in various professional fields. Established scientists can meet young scientists who may be recruited for a position in their organisation.

We look forward to seeing you all in Bern!

Organizing Committee:

Prof. Matthias Hamburger, PhD, Prof., SAPhS
matthias.hamburger@unibas.ch

Rudolf Brenneisen, PhD, Prof., SAPhS
rudolf.brenneisen@unibe.ch

Klaus Eyer, PhD, Prof., Aarhus University, SAPhS
eyerk@biomed.au.dk

Program

09:00 – 10:00 **Registration, Welcome Coffee**

10:00 – 10:15 **Welcome Addresses**

- Ursula von Mandach, PhD, Prof.
  Co-President SAPhS
- Verena Schröder, PhD, Prof.
  Co-President SAPhS

10:15 – 11:30 **Morning Session**

Chair: Verena Schröder, PhD, Prof.

10:15 – 10:45 **Lecture 1: New Paths to New Drugs**

Steve Pascolo, PhD, MD, PD
University Hospital Zurich

«Synthetic mRNA – New Paths to New Drugs»

10:45 – 11:15 **Lecture 2: Artificial Intelligence – Friend or Foe?**

Stéphane Guerrier, PhD, Prof.
University of Geneva

«Data Analytics and Artificial Intelligence in Pharmaceutical Sciences»

11:30 – 14:00 **Lunch Break and Poster Session**

Program (cont.)

14:00 – 16:15 **Afternoon Session**

Chair: Klaus Eyer, PhD, Prof.

14:00 – 14:45 **3 Short Oral Presentations of Selected Abstracts (SOPs)**

14:00 – 14:15 Ana Katrina Mapanao
PSI

«Preclinical radionuclide therapy of neuroendocrine neoplasms using radiolabeled somatostatin analogues»

14:15 – 14:30 Remo Eugster
University of Bern

«Machine learning-driven optimization of liposomal drug development in microfluidic systems»

14:30 – 14:45 Tamara Balsiger
University of Basel

«Natural product drug discovery pipeline reveals ellagic acid to reduce forgetting in *Caenorhabditis elegans* through specific Musashi inhibition»

14:45 – 15:15 **Lecture 3: Formulation Science and Manufacturing**

Jean-Christophe Leroux, PhD, Prof.
ETH Zurich

«3D Printing of Drug Eluting Devices:
A Research Perspective»

Program (cont.)

15:15 – 15:45	**Lecture 4: Improving Pharmaceutical Care**
	Carla Meyer-Massetti, PhD, Prof. University Hospital Inselspital Bern
	«Clinical Pharmacy – Improving Medication Safety»
15:45 – 16:15	**Coffee break**
16:15 – 16:45	**Roundtable**
	Moderator: Prof. Dr. Gerrit Borchard
	All speakers
	Discussion of Lectures 1-4
16:45 – 17:15	**Award Ceremony**
	SAPhS Fellow 2024
	Poster Prizes
17:15 – 17:30	**Closing Remarks**
17:30 – 18:30	**Farewell Apéro**

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Lectures

L-1

Lecture 1: New Paths to New Drugs

Steve Pascolo, PhD, MD, PD
University Hospital Zurich

«Synthetic mRNA – New Paths to New Drugs»

Biosketch:
Trained as an immunologist at the Pasteur Institute (Paris, France), Dr. Steve Pascolo used mouse models to test and develop mRNA-based vaccines (direct injection of mRNA) during his post-doc in Tübingen, Germany from 1998 till 2000.
In 2000, he co-founded CureVac with Dr. Hoerr and Dr. von der Mulbe. Dr. Pascolo was Chief Scientific Officer (CSO) of the company from 2000 until 2006, developing the technology, implementing the worldwide first GMP production of mRNA and performing the worldwide first clinical studies where humans (including himself in 2003) got injections of in vitro transcribed mRNA.
In 2006, he joined the oncology department of the University Hospital of Zurich, Switzerland and continued the development of immuno-therapies based on RNA. In 2008, he founded Miescher Pharma to support this work. In 2017, Dr. Pascolo implemented in Zurich an academic mRNA platform https://www.cancer.uzh.ch/en/Research/mRNA-Platform.html. In collaboration with several research and clinical departments in Zurich he optimizes, tests and implements mRNA based vaccines and therapies.

Lecture Abstract:
Although mRNA vaccines have been published in 1993 by a French team (Prof. Meulien, Paris), their development has been hindered by the prejudice associated to the supposed fragility of mRNA. Actually, mRNA is a very robust biomolecule (it can be heated up, frozen, lyophilized), easy to manufacture even at high scale and capable to lead to potent protein expression once administered naked or in particle formulations. Private investments in companies (for example CureVac created in 2000, BioNTech created in 2008 and Moderna created in 2011) have allowed to develop the potential of mRNA-based vaccines and therapies. The safety, versatility and efficacy of mRNA-based vaccines was evidenced during the COVID-19 pandemic: less than one year after the publication of the sequence of SARS-CoV-2, an mRNA vaccine against COVID-19 was approved and marketed. More mRNA vaccines (against infectious diseases and cancer) are in clinical developments and are expected to be approved in the coming years. In addition, mRNA-based therapies using non-immunogenic formulations of mRNA are being developed. Thus, the potential of synthetic mRNA in medicine is just starting to be unraveled and this versatile biomolecule is expected to be the active pharmaceutical ingredient in many future prophylactic and therapeutic drugs.

L-2

Lecture 2: Artificial Intelligence – Friend or Foe?

Stéphane Guerrier, PhD, Prof.
University of Geneva

«Data Analytics and Artificial Intelligence in Pharmaceutical Sciences»

Biosketch:
Stéphane Guerrier received the M.Sc. and B.Sc. degrees in environmental engineering from École Polytechnique Fédérale de Lausanne, and the Ph.D. degree in statistics from University of Geneva. He was an Assistant Professor with the Department of Statistics at The Pennsylvania State University (State College, PA, USA), and the Department of Statistics at University of Illinois at Urbana-Champaign (Champaign, IL, USA). Since January 2019, he has held an SNSF professorship position in Statistics and Data Science at University of Geneva. His research interests include biostatistics, computational statistics, signal processing, and data analytics.

Lecture Abstract:
As the pharmaceutical industry evolves, combining advanced statistical methods with Artificial Intelligence (AI) presents unprecedented opportunities to enhance research, development, and clinical practice. This talk will explore how traditional statistical techniques and emerging AI methodologies, particularly Machine Learning (ML), complement each other in pharmaceutical research.
Statistics has been the foundation for making inferences, testing hypotheses, and validating theories, ensuring that findings are robust and generalizable, thus supporting decision-making with controlled risk. However, ML adds a new dimension to our analytical toolkit, especially in predictive modeling and automating tasks like labeling data. While ML excels at making predictions, it must be used carefully, as even accurate predictions can be misleading if they are based on spurious correlations and fail to provide meaningful real-world insights.
One area where machine learning and AI show great potential is in analyzing Electronic Health Records (EHR). Due to the wide variation in coding systems across hospitals and countries, ML can automate and scale analyses that were previously impractical, enabling research collaboration across institutions and borders. Additionally, I will discuss the role of statistical transfer learning in using EHR data to solve related, data-scarce problems, thereby opening new avenues for research and improving the efficiency of processes like clinical trials.
In this talk, I will highlight both the promise and the challenges of incorporating ML into pharmaceutical research, advocating for a balanced approach that leverages the strengths of both statistics and ML to advance the field.

L-3

Lecture 3: Formulation Science and Manufacturing

Jean-Christophe Leroux, PhD, Prof.
ETH Zurich

«3D Printing of Drug Eluting Devices: A Research Perspective»

Biosketch:
Jean-Christophe Leroux
Institute of Pharmaceutical Sciences, Department of Chemistry and Applied Biosciences,
ETH Zürich, Zürich, Switzerland
E-mail: jleroux@ethz.ch

Jean-Christophe Leroux is a full professor of Drug Formulation and Delivery at the Institute of Pharmaceutical Sciences at the ETH Zurich since 2008. He was previously a Professor at the University of Montreal. He has made important fundamental and applied contributions to the fields of biomaterials and drug delivery and has been involved in the development of innovative bio-detoxification systems for the treatment of metabolite disorders. He is a fellow of the AAPS, EURASC, French Academy of Pharmacy, and the CRS, and the co-founder of the start-up pharmaceutical companies Versantis AG, Inositec AG and OBaris AG.

Lecture Abstract:
Three-dimensional (3D) printing is a potent method for creating affordable, personalized medical devices on demand (1). Among various 3D printing techniques, digital light processing (DLP) is notable for its fast fabrication of high-resolution objects. However, the production of bioresorbable medical devices using DLP is currently limited by the absence of appropriate biomedical inks. Here, we developed novel polyester-based inks that allow for DLP printing of therapeutic devices with tunable mechanical properties and degradation profiles. The most promising materials were used to design biodegradable customized airway stents. These stents degraded to soft innocuous hydrogels in vitro and disappeared 7 weeks after insertion in rabbits (2). The 3D printed stents could be loaded with drugs such as levofloxacin or nintedanib whose release kinetics could be adjusted by varying the copolymer composition. In addition, shape memory temperature-responsive stents were also 3D printed (3). By tuning the glass transition temperature of the polymeric matrix, it was possible to engineer folded stents that could be easily deployed at body temperature. Lastly, DLP was used as prototyping method to fabricate and optimize various oral formulations. Buccal suction patches mimicking the suction cup of octopuses were investigated for the administration of peptidic drugs (4) while pH-sensitive colonic capsules capable of encapsulating aqueous suspensions were investigated for the delivery of live probiotics. These studies open new perspectives for the rapid manufacturing of complex devices with superior properties.

Funding from SNSF (Sinergia CRSII5_177178 and 315230_197644) and ETH (Research grant 33-20-1) is acknowledged.

References
1) Liang K et al., Adv. Mater. 2019, 31, 1805680.
2) Paunovic N et al., Sci Adv 2021, 7, eabe9499.
3) Paunovic N et al. J Control Release 2023, 361, 417-426.
4) Luo Z et al., Sci Transl Med 2023, 15, eabq1887.

L-4

Lecture 4: Improving Pharmaceutical Care

Carla Meyer-Massetti, PhD, Prof.
University Hospital Inselspital Bern

«Clinical Pharmacy – Improving Medication Safety»

Biosketch:

BIOGRAPHICAL SKETCH

Carla Verena Meyer-Massetti

POSITION TITLE: Prof. Dr. phil. II, Assistant Professor of Clinical Pharmacy

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE	Completion Date	FIELD OF STUDY
University of Basel, Switzerland	MSc	2001	Pharmacy
Bruderholz Spital, Switzerland	FPH	2007	Hospital Pharmacy
University of California San Francisco, USA, and University of Basel, Switzerland	PhD	2011	Medication Safety
University of Basel, Switzerland	Postdoctoral studies	2020	Medication Safety in Home Care
Inselspital and University of Bern, Switzerland	Habilitation	2024	Clinical Pharmacy

Positions and Honors

Currently, I'm working as an assistant professor of clinical pharmacy in the Department of General Internal Medicine at the University Hospital of Bern and I'm an adjunct researcher at the Institute for Primary Care BIHAM, University of Bern. Since many years, I'm active as an associated researcher in the Pharmacoepidemiology and Clinical Pharmacy group of Prof. Christoph Meyer at the University of Basel, where I pursue several ongoing research projects with nursing science, mainly focusing on long-term care and home care.

My current position is funded by pharmaSuisse, and my research has also been supported during the last few years by the LOA fund, the Canton of Lucerne, Home Care Lucerne, the Kollegium für Hausarztmedizin, the ZUW Center for continuous education at the University of Bern as well as smarter medicine.

Motivation and Activities

In my role as an educator in the new master's curriculum at the University of Bern since 2020, I have a chance to teach our students about evidence-based clinical pharmacy practice in hospitals, home care, long-term care, hospice care and community pharmacy. I'm very involved in establishing interprofessional pre-grad courses for pharmacy, medicine and nursing students, where implementing evidence-based practice is a core topic.

Since October 2023, I'm also organzing the interprofessional CAS Medication Safety at the University of Bern.

In my clinical work at Inselspital, I develop and implement new, evidence-based clinical pharmacy services that also allow me to collect and publish data to share with other colleagues, interested in doing the same. My extensive network has allowed me to collaborate internationally in a wide array of projects, which make my workdays very interesting.

Contributions to Science

Research shows clearly, that pharmacists in interprofessional teams can improve medication safety for our patients. However, in many settings, they are not yet an integral part of a patient's care team.

Because resources are limited, my research focuses on effective approaches to clinical pharmacy practice, prioritizing vulnerable patients. Aside from 3 PhD students, I have mentored more than 50 master's theses in pharmacy, nursing sciences and public health, focusing on medication safety research in home care, long-term care, palliative care and transition of care. Some of our recent publication are displayed below:

Goetschi AN, Verloo H, Wernli B, Wertli MM, Meyer-Massetti C. Prescribing pattern insights from a longitudinal study of older adult inpatients with polypharmacy and chronic non-cancer pain. Eur J Pain. 2024.

Möckli N, Simon M, Denhaerynck K, Trutschel D, Martins T, Meyer-Massetti C, Zúñiga F. How external and agency characteristics are related to coordination in homecare - findings of the national multicenter, cross-sectional SPOTnat study. BMC Health Serv Res. 2024.

Schönenberger N, Blanc AL, Hug BL, Haschke M, Goetschi AN, Wernli U, Meyer-Massetti C. Developing indicators for medication-related readmissions based on a Delphi consensus study. Res Social Adm Pharm. 2024.

Wernli U, Hischier D, Meier CR, Jean-Petit-Matile S, Kobleder A, Meyer-Massetti C. Pharmacists' clinical roles and activities in inpatient hospice and palliative care: a scoping review. Int J Clin Pharm. 2023.

Favez L, Zúñiga F, Meyer-Massetti C. Exploring medication safety structures and processes in nursing homes: a cross-sectional study. Int J Clin Pharm. 2023.

Gnägi R, Brunkert T, Zúñiga F, Meyer-Massetti C. Development of a medication literacy assessment instrument (MELIA) for older people receiving home care, J Adv Nurs. 2022.

Lecture Abstract:

Medication related problems - consisting of adverse drug reactions and medication errors – are among the most prevalent adverse events in health care. While we know a lot about their types and origin, little is known about how two optimize medication safety sustainably.

This is a challenge and opportunity for clinical pharmacists in different healthcare settings.

In this talk, the Clinical Pharmacy team of the Department of General Internal Medicine at the Inselspital Bern would like to present the current status of medication safety in Switzerland and follow a fictitious patient on her journey through our health care system. Different settings, in ambulatory, acute inpatient and longterm care, present different challenges to the optimization of medication safety and ultimately patient safety.

We will focus on especially vulnerable patients, their prioritization for medication reconciliation and medication review and the inclusion of patients and caregivers into the healthcare team in the scope of our research that we translate directly to patient care and teaching. Our research shows, that pharmacists can contribute significantly to medication safety in an interprofessional team.

Posters

I. PHARMACEUTICAL BIOLOGY / PHYTOPHARMACOLOGY

P-I-1 **C. Steuer:** Chemical and enantiomeric stability of terpenes commonly found in *Cannabis sativa* L. flowers
P-I-2 **E. Gezer:** Marine actinobacterium as a source of natural products against multiple myeloma
P-I-3 **A. Zingg**: Insights into the mechanism of action of a withanolide derivative in multiple myeloma
P-I-4 **T. Balsiger:** Natural product drug discovery pipeline reveals ellagic acid to reduce forgetting in *Caenorhabditis elegans* through specific Musashi inhibition
P-I-5 **L. Höing:** Biosynthesis of the bacterial siderophoric antibiotic 3,7-dihydroxytropolone through enzymatic salvaging of catabolic shunt products
P-I-6 **R. Han:** Screening for natural products inhibiting PI3K/AKT pathways in melanoma: investigation of *Mammea americana* EtOAc extract
P-I-7 **H.G. Weddeling:** Target identification for rubromycin antibiotics and their bioengineering via late-stage tailoring enzymes
P-I-8 **M. Rakhmanov:** More than detoxification: How marine bacteria utilize glutathione to produce sulfur containing antibiotics

II. PHARMACEUTICAL TECHNOLOGY

P-II-1 **R. Eugster**: Machine learning-driven optimization of liposomal drug development in microfluidic systems
P-II-2 **Ch. Greitens:** Intracellular localization of a protein-based DNA delivery system
P-II-3 **H. Krupke:** Characterization of a biodegradable suction patch for transbuccal peptide delivery
P-II-4 **S. Honrath:** Proteomics-supported improvement of TFAMoplex-mediated transfection
P-II-5 **J. Kalasová:** Developing a novel solubility measurement technique based on second harmonic generation
P-II-6 **V. Patrulea:** Physics versus chemistry: Innovations in cryopreservation for cell-based therapies via ultrasonication or chemical modification
P-II-7 **I. Bortolazzo:** Lipid-degrading enzymes as permeation enhancers
P-II-8 **S. Teworte:** Modelling fibrosis in endometriosis using a 3D cell culture system based on fibronectin (FN)-silk
P-II-9 **D. Gao:** Heat application for enhanced oral peptide absorption – a proof-of-concept
P-II-10 **R. Gazzi:** 3D-printed lipid mesophases for the treatment of chronic liver disease

III. PHARMACOEPIDEMIOLOGY

P-III-1 **D. Zappalà:** Market uptake and pattern of use of emerging topical rosacea medications in Switzerland: A descriptive study using Swiss claims data
P-III-2 **M. Rainer:** Development of a real-time opioid monitoring system at a Swiss cantonal hospital: An observational study
P-III-3 **N.A. Stohler:** Uptake of pertussis vaccine during pregnancy following guideline changes in Switzerland: A descriptive study using Swiss claims data
P-III-4 **K. Rekk:** Are specific DOACs favored by clinicians for patients with atrial fibrillation post stroke? A secondary data analysis of the MAAESTRO-study in Basel, Switzerland

P-III-5 **N.L. Wittwer:** The importance of assessing drug-drug-gene interactions
P-III-6 **M. Roth:** Antidepressant drug switching in the Swiss population with a focus on Escitalopram and pharmacogenetically relevant drugs: A drug utilization study using the Helsana Database
P-III-7 **L. Velez-Nandayapa:** Characteristics of patients using anti-glaucoma therapy in the UK, 2000-2022
P-III-8 **I. Muzzarelli:** Are potentially inappropriate mediations (PIMs) associated with rehospitalisation and death within three months? A systematic review and meta-analysis

IV. CLINICAL PHARMACY / CLINICAL PHARMACOLOGY

P-IV-1 **L. Solh Dost:** Identifying current practices and areas for improvement in medication management during care transition through an Interprofessional Collaboration Framework
P-IV-2 **N. Schönenberger:** Medication reconciliation at hospital admission: findings and learnings from a pilot study on a Swiss general internal medicine ward
P-IV-3 **A.N. Goetschi:** Quality indicators for the pharmacological management of chronic non-cancer pain in older adult patients: an integrative review
P-IV-4 **C. Godot:** Digital healthcare services in community pharmacies: The PneumoscopeTM case study
P-IV-5 **P. Stoyanova:** PharmVisit: Interprofessional ward rounds with clinical pharmacist have the potential to improve medication safety and foster interprofessional collaboration
P-IV-6 **M.C. Sarbach:** Personalized adherence interventions with electronic monitoring: A case report on polypharmacy management in epilepsy

V. MOLECULAR PHARMACOLOGY / MOLECULAR MEDICINE

P-V-1 **A. Blagojevic:** From parasite to therapeutic: Expression, functional characterization, and therapeutic potential of the leech-derived complement inhibitor gigastasin
P-V-2 **P. Rüthemann:** squeezeMD: An open-source tool for protein interaction visualization and analysis
P-V-3 **F. Meyer:** Development of a platform for the expression of recombinant antibodies targeting complement proteins
P-V-4 **S. Vogt:** Breaking the boundaries of the clinical C3 inhibitor compstatin: Development of species-tolerant and long-acting analogs
P-V-5 **Z.V. Kürsteiner:** A DNAzyme's catalytic comeback: Why two deleterious mutations are better than one

VI. PHARMACOLOGY / BIOPHARMACY

P-VI-1 **K. Krieger:** Tolerability of [161Tb]Tb-SibuDAB, a novel radioligand for the treatment of prostate cancer, in healthy mice
P-VI-2 **A.K. Mapanao:** Preclinical radionuclide therapy of neuroendocrine neoplasms using radiolabeled somatostatin analogues
P-VI-3 **C. Vaccarin:** Development of a novel transthyretin-binding PSMA radioligand characterized by an improved biodistribution profile
P-VI-4 **M. Rysz:** Distribution and metabolic study in SLCO2B1+/+ and Slco2b1 -/- rats receiving chronic oral treatment of erlotinib
P-VI-5 **M. Karpouchtsi:** Mission (im)possible: Target identification for natural products inhibiting oncogenic ERK and AKT signaling pathways in melanoma
P-VI-6 **F. Brigger:** Biochemical characterization of skin mitochondrial extracellular vesicles
P-VI-7 **N. Paloumpis:** Deciphering the influence of SLCO2B1 on CYP3A-function: a rat study using midazolam

P-VI-8 **L. Potzel:** *In vitro* analysis of Cytochrome P450 induction and its impact on coproporphyrin levels

P-VI-9 **N. Vavilthota:** Investigating the dual functionality of branched tetrameric peptides for their applications in chronic wound treatment

P-VI-10 **J. Vérièpe-Salerno:** Modulation of autophagy against multiple myeloma and use of *C. elegans* as a study tool

I. PHARMACEUTICAL BIOLOGY / PHYTOPHARMACOLOGY

P-I-1

Chemical and enantiomeric stability of terpenes commonly found in *Cannabis sativa* L. flowers

J. Raeber[1], B. Bajor[1], M. Poetzsch[2], C. Steuer[1]

[1]Departement of Chemistry and Applied Biosciences, Pharmaceutical Analytics, ETH Zurich, 8093 Zurich
[2]Swiss Drug Testing GmbH, Technoparkstrasse 2, CH-8406 Winterthur

Introduction: *Cannabis sativa* L. may be the most discussed medicinal plant in recent years. With the increasing commercialization of *C. sativa* L.-based products for medicinal use and recreational purposes, there is a growing need for well-controlled products [1,2]. While the stability of main cannabinoids, such as THC and CBD, is well understood, data on the chemical and enantiomeric stability of terpenes remain scarce in the literature. This is even though terpenes possess individual pharmacological properties and may contribute to the overall therapeutic effectivity of *C. sativa* [3,4].

Aims: The present work focuses on the impact of environmental stresses, such as UV light and temperature, on terpene patterns in cannabis flowers and extracts thereof. Obtained data is valuable for biomarker discovery of aging processes. Furthermore, we suggest closely evaluating the enantiomeric ratio of terpenes in cannabis since this could significantly influence the proposed synergy and resolve conflicting results of the entourage effect. This comprehensive understanding will support future cannabis pharmacology studies and may encourage the use of standardized cannabis extracts. Finally, we aim to improve quality control and increase patient safety and drug efficacy.

Methods: For comprehensive monitoring of terpene patterns, four gas chromatographic methods based on polar, apolar, and chiral stationary phases combined with either FID or MS detection were developed and validated according to international guidelines. These methods successfully separated and quantified 29 terpenes, including 13 enantiomers and 5 diastereomers specific to *C. sativa*. Furthermore, authentic *C. sativa* L. flowers, and ethanolic extracts were subjected to UV light and heat treatments to detect time-dependent degradation reactions.

Results: Mono- and sesquiterpenes generated a unique pattern of degradation products, resulting in a diverse array of oxidation and cyclization products. p-Cymene was identified as a significant product of terpene aging. No enantiomeric conversion was observed, suggesting that (-)-α-pinene formation in cannabis extracts, for example, originates from other terpenes.

Conclusions: Terpenes have different degradation rates despite being structurally similar. In addition, different enantiomeric ratios were observed in *C. sativa* depending on cultivar and growth conditions. We conclude that the enantiomeric terpene pattern is species-specific and must be considered for therapeutic applications.

Keywords: Terpenomics, stability testing, chiral terpene pattern analysis, *Cannabis sativa* L.

References:

[1] Raeber, J.; Poetzsch, M.; Schmidli, A.; Favrod, S.; Steuer, C. Simultaneous quantification of terpenes and cannabinoids by reversed-phase LC-APCI-MS/MS in Cannabis sativa L. samples combined with a subsequent chemometric analysis. Anal Bioanal Chem. 2024. DOI: https://10.1007/s00216-024-05349-y.
[2] Zivovinovic, S.; Alder, R.; Allenspach, M. D.; Steuer, C. Determination of cannabinoids in Cannabis sativa L. samples for recreational, medical, and forensic purposes by reversed-phase liquid chromatography-ultraviolet detection. Journal of Analytical Science and Technology. 2018, 9 (1), 27. DOI: https://10.1186/s40543-018-0159-8.
[3] Russo, E. B. Taming THC: potential cannabis synergy and phytocannabinoid-terpenoid entourage effects. Br J Pharmacol. 2011, 163 (7), 1344-1364. DOI: https://doi.org/10.1111/j.1476-5381.2011.01238.x.
[4] Allenspach, M.; Steuer, C. α-Pinene: A never-ending story. Phytochemistry. 2021, 190, 112857. DOI: https://doi.org/10.1016/j.phytochem.2021.112857.

P-I-2

Marine actinobacterium as a source of natural products against multiple myeloma

E. Gezer[1,2], E. Bilgi[3], E. Bedir[3], M. Cuendet[1,2]

[1] *School of Pharmaceutical Sciences, University of Geneva, 1211 Geneva*
[2] *Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, 1211 Geneva*
[3] *Department of Bioengineering, Faculty of Engineering, Izmir Institute of Technology, 35430, Urla, Izmir, Turkey*

Introduction: Multiple myeloma is a hematologic malignancy characterized by the abnormal proliferation of clonal plasma cells. Patients frequently experience relapse and develop resistance to the currently available drugs. Therefore, there is a need to find new treatment options, and the investigation of sources of natural products is of interest [1].
Aims: The present study aimed at investigating the effects of various fermentation conditions on the production of secondary metabolites in *Streptomyces cacaoi*. Additionally, the antiproliferative activity of compounds produced under conditions that were previously optimized based on compound diversity and extract antiproliferative activity, was evaluated.
Methods: Response Surface Methodology (RSM) with a Box-Behnken design was used to identify optimal fermentation conditions. The antiproliferative activity of the ethyl acetate extracts was tested in RPMI 8226 multiple myeloma cells at a concentration of 45 µg/mL and evaluated as a response in the experimental design. The produced compounds were isolated and structurally elucidated after large-scale fermentation under optimized conditions. The antiproliferative activity of the compounds was determined against the multiple myeloma cell lines RPMI 8226 and KMS-12-BM, as well as their drug-resistant counterparts.
Results: The extracts, obtained based on the Box-Behnken design, showed antiproliferative activities at varying potencies. While the most active extracts inhibited the proliferation of RPMI 8226 cells by 100% at a concentration of 45 µg/mL, some other extracts were not active (<30%). Water type (distilled *vs* seawater), glycerol, peptone, and $CaCO_3$ concen-trations significantly affected the production of bioactive compounds. Strong interactions were observed between some factors. For example, seawater enabled the production of more active extracts at a lower temperature, but no significant differences were observed between seawater and distilled water at a higher temperature. Similarly, $CaCO_3$ significantly increased the production of active extracts when the fermentation was conducted in distilled water, but not in seawater. Finally, thirteen polyether-type polyketides were isolated from an extract obtained with optimized conditions. The major compound was characterized as K-41A, and eleven compounds were identified as methylated/demethylated and glycosylated/ deglycosylated derivatives of K-41A [2]. Three compounds showed antiproliferative activity with IC_{50} values in the low micromolar range against four multiple myeloma cell lines.
Conclusion: This study demonstrates the critical impact of fermentation conditions on producing bioactive natural products against multiple myeloma cancer cell lines.

Keywords: Multiple myeloma*, Streptomyces cacaoi*, optimization, fermentation, cytotoxicity

References:
[1] Yu CC, Li Y, Cheng ZJ, Wang X, Mao W, Zhang YW. Front Pharmacol 2022; 13: 818179.
[2] Gezer E, Üner G, Küçüksolak M, Kurt MÜ, Doğan G, Kırmızıbayrak PB, Bedir E. Phytochemistry 2022; 195: 113038.

P-I-3

Insights into the mechanism of action of a withanolide derivative in multiple myeloma

A. Zingg[1,2,3], K. Wijeratne[4], L. Gunatilaka[4], M. Cuendet[1,2,3]

[1] *School of Pharmaceutical Sciences, University of Geneva, 1211 Geneva*
[2] *Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, 1211 Geneva*
[3] *Translational Research Centre in Oncohaematology, 1211 Geneva*
[4] *Southwest Center for Natural Products Research, School of Natural Resources and the Environment, College of Agriculture, Life and Environmental Sciences, The University of Arizona, Tucson, Arizona 85719, United States*

Introduction: Multiple myeloma arises from the clonal proliferation of plasma cells in the bone marrow. Despite the availability of a wide range of drugs, current multiple myeloma treatments are not sufficient to cure patients who often relapse and develop drug resistance. Natural products can be used as a source of inspiration to find new drugs thanks to their structural diversity. A previous structure-activity relationship study on withanolides, which are naturally occurring steroidal lactones, led to the identification of a compound named C13.
Aim: This work aimed to assess the biological activity and the underlying mechanism of C13.
Methods: The antiproliferative activity was measured using the XTT assay after 72 h treatment. Patient plasma cells were isolated from bone marrow aspirate using a CD138+ marker and cytotoxicity was measured after 24 h treatment by flow cytometry using Draq7. The anti-angiogenic activity was evaluated after 48 h treatment through a 3D angiogenesis *in vitro* model. This model allows endothelial vessels to form in a fibrin gel, stimulated by angiogenic factors produced by an upper fibroblast layer. Drug combinations were tested for 72 h, and the combination index was calculated with the Chou-Talalay method using the CompuSyn software. 3D co-culture spheroids were formed with RPMI 8226, mesenchymal stem cells, and endothelial progenitor cells in a 2:1:1 ratio and treated for 48 h. Proteomic analysis was performed after 12 h treatment of RPMI 8226 cells.
Results: To evaluate the antiproliferative activity of C13 in a heterogenous disease such as multiple myeloma, patients' primary malignant plasma cells were used. After 24 h treatment, C13 at 500 nM displayed a similar activity than in the multiple myeloma cell line RPMI 8226 with a percentage of cell viability of 28% compared to the negative control (100%). To assess C13 activity in a more representative model, 3D co-culture spheroids were used. C13 antiproliferative activity was in the high nM range with an IC_{50} of 342 nM. As anti-cancer drugs are mainly given in combinations, the antiproliferative activity of C13, combined with clinically used drugs, was evaluated. Synergistic effects were observed when combining C13 (25 nM), dexamethasone (8 nM), and selinexor (12.5 nM) in RPMI 8226 cells (CI = 0.34 ± 0.09) with high antiproliferative activity (91.5 ± 3.6%). The impact of C13 on angiogenesis, a major tumor-supporting process in the bone marrow microenvironment, was studied. A decrease in the vessel network of about 30% was observed after treatment with C13 at 160 nM compared to the control. The mechanism behind C13 antitumoral activity was then investigated. Proteomic analysis, confirmed by western blot, showed a dose-dependent increase of p21 and a decrease of the cell cycle regulators cyclin D1, cyclin-dependent kinase 4, and cyclin-dependent kinase 6. C13 activity was evaluated in other hematological cancer cell lines (THP-1, HL-60, Jurkat, Jijoye, and Granta-519). IC_{50} values ranged from 24 to 44 nM which are comparable to results found in RPMI 8226 (IC_{50} values of 22 nM).
Conclusion: Based on the above data, the upstream molecular pathways and targets of C13 against multiple myeloma warrant a detailed investigation.

Keywords: Multiple myeloma, natural product, hit compound, mechanistic studies, micro-environment

P-I-4

Natural product drug discovery pipeline reveals ellagic acid to reduce forgetting in *Caenorhabditis elegans* through specific Musashi inhibition

T. Balsiger[1], A. Stetak[2,3], R. Hagmann[1], K. Huynh[2], P. Solis[4], M. Hamburger[1], A. Papassotiropoulos[2,3], R. Teufel[1], and E. Garo[1]

[1] *Department of Pharmaceutical Sciences, University of Basel, 4056 Basel*
[2] *Research Cluster Molecular and Cognitive Neurosciences, Department of Biomedicine, University of Basel, 4056 Basel*
[3] *University Psychiatric Clinics, University of Basel, 4056 Basel*
[4] *Center for Pharmacognostic Research and Panamanian Flora (CIFLORPAN), Faculty of Pharmacy, University of Panama, 0801 Panama City*

Introduction: Memory maintenance and forgetting are fundamental processes in our lives. However, pathological forgetting as in neurodegenerative diseases (e.g. Alzheimer`s disease) leads to serious cognitive impairment. Recently, Musashi (MSI), an RNA-binding protein and translational regulator, has emerged as a crucial player in promoting forgetting [1]. The constantly aging population and the lack of treatment against forgetting, prompted us to search for new MSI inhibitors.
Aim: Discovery of new lead compounds from plant origin inhibiting human MSI.
Methods: Our in-house library of 2`576 plant extracts was screened for MSI inhibition using a fluorescence polarization assay. HPLC-based activity profiling, which links activity to distinct peaks in an extract, was used to select the most promising hits, such as the MeOH extract of *Freziera candicans* Tul., which was then scaled up for isolation. All isolated compounds were first tested *in vitro*, and the most active compound ellagic acid (EA) was then further subjected to *in vivo* short- and long-term associative memory studies in *Caenorhabditis elegans* (*C. elegans*) [2]. Moreover, the impact of MSI inhibition through genetic deletion or EA treatment on Aβ & tau Alzheimer *C. elegans* strains was investigated by chemotaxis index, motility, and brood size experiments.
Results & Conclusion: A total of 11 active natural products from *Freziera candicans* Tul. were isolated, inhibiting human MSI in the low micro- to nanomolar range *in vitro*. The most active compound was identified as EA, a polyphenol common in many foods such as nuts and berries. EA significantly improved short- and long-term associative memory in *C. elegans*, which was further confirmed to be mediated through specific MSI inhibition. Genetic deletion of MSI in the Aβ & tau Alzheimer strains rescued all three tested phenotypes and similar effects were achieved with EA treatment. Additional experiments will include extended investigations of EA binding to MSI by surface plasmon resonance and X-ray crystallography.

Keywords: Forgetting, Alzheimer, natural products, Musashi inhibitors, ellagic acid, *C. elegans*

References:

[1] Hadziselimovic N, Vukojevic V, Peter F, Milnik A, Fastenrath M, Fenyves B, Hieber P, Demougin P, Vogler C, De Quervain D, Papassotiropoulos A, Stetak A. Forgetting is regulated via musashi-mediated translational control of the Arp2/3 complex. Cell 2014; 156: 1153–1166
[2] Mastrandreas P, Arnold A, Boglari C, De Quervain D, Stetak A, Papassotiropolous A. (-)- Gossypol Inhibition of Musashi-Mediated Forgetting Improves Memory and Age-Dependent Memory Decline in Caenorhabditis elegans. Mol Neurobiol 2023; 60: 820–835

P-I-5

Biosynthesis of the bacterial siderophoric antibiotic 3,7-dihydroxytropolone through enzymatic salvaging of catabolic shunt products

L. Höing[1], S. Sowa[1], M. Toplak[2], J. Reinhardt[1], R. Jakob[3], T. Maier[3], M. Lill[4], R. Teufel[1]

[1] *Pharmaceutical Biology, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel*
[2] *Bioorganic Chemistry, Biocatalysis, University of Graz, 8010 Graz, Austria*
[3] *Biozentrum, University of Basel, 4056 Basel*
[4] *Computational Pharmacy, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel*

Introduction: Tropolones are a group of natural products with potent metal-chelating properties that exhibit antibacterial, antiviral and antitumoral activity [1]. These compounds are hydroxylated derivatives of tropone that consists of a seven-membered, non-benzenoid, aromatic carbon-ring with an additional keto-group. In bacteria the precursor for these compounds surprisingly originates from primary metabolism, i.e. the CoA-dependent catabolism of phenylacetic acid (PAA). However, depending on the producing strain, different sets of enzymes are used to modify this precursor. For the gram-positive *Streptomyces sp.*, the gene cluster encoding the dihydroxytropolone biosynthetic machinery was identified by gene-knockout studies [2].
Aims: In this study we investigated dihydroxytropolone formation in *Streptomyces sp.* by *in vitro* reconstitution of the biosynthetic pathway and elucidated the role of involved enzymes. Furthermore, we identified and investigated another enzyme, which interacts with (di)hydroxy-tropolone and presumably plays an important part in making iron accessible for the organism.
Methods: Heterologously produced enzymes were used to gain insight into the individual biosynthetic steps and reaction mechanisms of the partaking enzymes. Conducted assays were analyzed via LC-MS, accumulating products were compared to chemically synthesized standards. Structure of key enzymes was analyzed by X-ray crystallography.
Results: Accordingly, the CoA-ester bond from the precursor molecule originating from PAA catabolism gets cleaved off. In an unanticipated series of reactions comprising hydroxylation, decarboxylation and ring oxidation tropolone gets formed. This compound undergoes two consecutive ring-hydroxylations and is finally transformed to dihydroxytropolone. A crystal structure from one of the key enzymes could be obtained, which gave hints about the underlying reaction mechanism [3]. Another enzyme in the same gene-cluster, a siderophore-interacting protein (SIP) is shown to interact with (di)hydroxytropolone. Its presumed role of reducing iron-III bound to (di)hydroxytropolones and making it therefore accessible for the organism is currently being investigated. An obtained crystal structure is giving additional insights into possible reaction mechanisms.
Conclusions: The biosynthesis pathway for dihydroxytropolone could be reconstituted successfully *in vitro*. Taken together, the discovered enzyme functionalities substantially differ from the previously proposed roles that were based on gene knock out studies. Currently, the mechanism of an additional enzyme, which employs the metal-chelating properties of (di)hydroxytropolones to make iron accessible for the organism, is investigated in more detail. These insights could help to engineer more potent antibiotics (coupled to siderophores) in the future.

Keywords: Tropolone, secondary metabolites, antibiotics, siderophores, *Streptomyces sp.*

References:
[1] Duan Y et al. ChemBioChem 2020; 21: 2384-2407
[2] Chen R et al. Appl Environ Microbiol 2018; 84: e01321-18
[3] Höing L et al. (2024), Chem Sci 2024; 15: 7749-7756

P-I-6

Screening for natural products inhibiting PI3K/AKT pathways in melanoma: investigation of *Mammea americana* EtOAc extract

R. Han[1], T. Balsiger[1], M. Dobrzyński[2], J. K. Reinhardt[1], L. Dürr[1], T. Hell[1], P. Solis[3], M. Hamburger[1], O. Pertz[2], R. Teufel[1], E. Garo[1]

[1] University of Basel, Department of Pharmaceutical Sciences, Klingelbergstrasse 50, 4056 Basel
[2] University of Bern, Institute of Cell Biology, Baltzerstrasse 4, 3012 Bern
[3] University of Panama, Faculty of Pharmacy, PA-0801 Panama City

Introduction: Malignant melanoma is the deadliest skin cancer, and its incidence has risen significantly over the past decades. Mutations are frequently observed in the PI3K/AKT and MAPK/ERK pathways, leading to abnormal cell proliferation. Targeted therapy of these pathways showed spectacular initial results, but drug resistance appears after a few months only. Combination therapy has shown improvement in the progression-free and overall patient survival, and additional compounds are urgently needed to complete the arsenal of available drugs.
Aims: Finding new AKT/ERK pathway inhibitors in melanoma.
Methods: Our in-house library of 2'576 crude extracts was screened by using an innovative high-content screening (HCS) assay to quantify AKT and ERK signaling in melanoma cells [1]. The hits were submitted to the HPLC-based activity profiling approach for further prioritization.
Results: The EtOAc extract from *Mammea americana* leaves, one of the promising hits, was scaled-up and led to the isolation of ten new betulinic acid derivatives and nine known coumarins. The coumarin theraphin B was the most active compound with an IC_{50} of 37 ± 0.4 µM on the PI3K/AKT pathway. Interestingly, although betulinic acid has been reported to have anti-tumor activity in melanoma mice model [2], the betulinic acid analogues did not show any inhibition of AKT and ERK, suggesting a different mechanism, unrelated to the AKT signaling pathway, to explain its *in vivo* activity.
Conclusions: This phytochemical investigation of *Mammea americana* EtOAc extract, resulted in the isolation of nineteen compounds, including ten previously undescribed triterpenes and nine coumarins. Theraphin B was inhibiting PI3K/AKT signaling pathway with an IC_{50} value of 37.0 ± 0.4 µM.

Keywords: *Mammea americana*, melanoma, high-content screening, coumarin, ECD

References:

[1] Dürr L, Hell T, Dobrzyński M, Mattei A, John A, Augsburger N, Bradanini G, Reinhardt JK, Rossberg F, Drobnjakovic M, Gupta MP, Hamburger M, Pertz O, Garo E. High-content screening pipeline for natural products targeting oncogenic signaling in melanoma. J Nat Prod 2022; 185(4): 1006-1017.
[2] Pisha E, Chai H, Lee I, Chagwedera TE, Farnsworth NR, Cordell GA, Beecher CWW, Fong HHS, Kinghorn AD, Brown DM, Wani MC, Wall ME, Hieken TJ, Gupta TKD, Pezzuto JM. Discovery of betulinic acid as a selective inhibitor of human melanoma that functions by induction of apoptosis. Nature Med 1995; 1: 1046-1051.

P-I-7

Target identification for rubromycin antibiotics and their bioengineering *via* late-stage tailoring enzymes

H.G. Weddeling, S.T. Sowa, F. Röthlin, R. Teufel

Department of Pharmaceutical Sciences, University of Basel, 4056 Basel

Introduction: Rubromycins are a class of bacterial aromatic polyketides with antimicrobial and cytotoxic activities. Moreover, rubromycins are known to inhibit proteins that typically interact with nucleic acids, albeit the true molecular targets likely remain elusive [1]. Interestingly, they feature an unusual [5,6]-spiroketal moiety, disrupting their structural planarity. After the initial formation and cyclization of the linear polyketide, various late-stage tailoring enzymes like oxygenases, reductases and transferases finalize the biosynthesis. Their potent bioactivities and structural diversity make rubromycins particularly interesting to study. Among the rubromycins, hyaluromycin stands out with its unusual 2-amino-3-hydroxycyclopent-2-enone (C_5N) moiety, which presumably increases its water solubility and possibly pharmacokinetic properties [2]. As the involved amide synthetase has homologues with high substrate promiscuity, this enzyme harbors distinct biotechnological potential to further diversify and pharmacologically improve the rubromycin structural family [3].
Aims: Elucidation of late-stage tailoring steps during hyaluromycin biosynthesis and bioengineering of C5N-containing rubromycin analogs, as well as identification of additional molecular targets.
Methods: The potentially involved genes have been cloned from *Streptomyces hyaluromycini* and heterologously expressed in *Escherichia coli*. An alternative rubromycin acid substrate has been produced by purification and basic hydrolysis of purpuromycin from *Actino-planes ianthinogenes* cultures. Enzymes were characterized using biophysical methods and by *in vitro* assays with subsequent elucidation of reaction products *via* LC-MS, LC-HRMS/MS and NMR.
Results: The involved 5-aminolevulinate synthase and acyl-CoA ligase could be successfully combined with the amide bond synthetase from *S. hyaluromicini* to form the C_5N adduct of purpuromycin acid *in vitro*. The reaction could be optimized regarding the buffer and pH, as well as prevention of allosteric inhibition of the amide bond synthetase, thereby increasing turnover substantially. Additionally, initial alternative amide derivatives have been generated with the rubromycin backbone. Furthermore, strategies are currently being employed for target identification.
Conclusion: The successful reconstitution of the late-stage tailoring steps during hyaluromycin biosynthesis prompts us to further investigate the biotechnological potential of the involved enzymes. A mutagenesis approach to increase the promiscuity of the amide synthetase regarding its amine substrate will be used with the aim of producing a multitude of «unnatural» rubromycin analogs. This diversification of the rubromycin-type structural family facilitates the development towards new antibiotics and anti-cancer agents.

Keywords: Rubromycin, aromatic polyketides, PKS, antibiotics, enzymology, *Streptomyces*

References:
[1] Lin P et al. Isolation, biosynthesis, and biological activity of rubromycins derived from actinomycetes. Eng Microbiol 2022; 2: 100039.
[2] Harunari E et al. Hyaluromycin, a New Hyaluronidase Inhibitor of Polyketide Origin from Marine Streptomyces sp. Mar Drugs 2014; 12: 491–507.
[3] Tang Q et al. Broad Spectrum Enantioselective Amide Bond Synthetase from Streptoalloteichus hindustanus. ACS Catal 2024; 14: 1021–1029.

P-I-8

More than detoxification: How marine bacteria utilize glutathione to produce sulfur-containing antibiotics

M. Rakhmanov, S. Sowa, T. Martelli, J. Felsch, R. Teufel

Department of Pharmaceutical Sciences, University of Basel, 4056 Basel

Introduction: Almost all domains of life have developed glutathione (GSH) biochemistry as a major way of dealing with potentially harmful metabolic dead-ends, xenobiotics and oxidative stress. While well studied in eukaryotes, e.g. human drug metabolism, its role in bacteria received far less attention but has at the same time emerged to be more complex than previously assumed [1]. Bioinformatic analyses of bacterial genomes unveiled that GSH-conjugation might play a crucial role in the biosynthesis of secondary metabolites, one example being the structurally unique marine antibiotic tropodithietic acid (TDA), which contains two sulfur atoms that appear to be incorporated *via* non-canonical ways. With antibiotic resistance globally on the rise, investigating unusual and underexplored antibiotics such as TDA is of imminent importance.
Aim: Deciphering the cryptic sulfur incorporation during TDA biosynthesis with the aim to generate novel antibiotics using the underlying biochemistry.
Methods: Enzymes involved in TDA production were chosen based on bioinformatic predictions and knockout studies. These candidate enzymes were heterologously produced, and their biochemical functions assessed using *in vitro* assays as well as biophysical methods. To test biosynthetic hypotheses structural analogues of postulated intermediates were synthesized and used as surrogate substrates for these investigations. Complementary structural studies were conducted using X-ray crystallography to scrutinize the underlying enzymology.
Results: We were able to successfully reconstitute a three-enzyme cascade *in vitro*, leading to the integration of a thiol group into a model substrate. More precisely, it could be shown that a GSH moiety attached to an aromatic backbone *via* a C-S bond is sequentially degraded by a γ-glutamyl-cyclotransferase, dipeptidase and cysteine-S-conjugate-β-lyase. Additionally, the structure of the β-lyase could be elucidated using X-ray crystallography, granting insight into the catalytic mechanism behind the C-S bond cleavage.
Conclusion: These findings corroborate the notion of GSH as the source of sulfur in TDA and pave the way for the biotechnological production of analogous compounds. Interestingly, the degradation of GSH to a reactive thiol has been described in human drug and xenobiotic metabolism before. However, it seems to be a metabolic end-product associated with nephrotoxicity [2]. It appears that bacteria have found a way to leverage this reactivity, turning a challenge into an opportunity.

Keywords: Antibiotics, bacterial metabolism, enzymes, glutathione, natural products

References:
[1] Vuilleumier S. Bacterial Glutathione S-Transferases: What Are They Good For? J Bacteriol 1997; 179 (5): 1431–1441. https://doi.org/10.1128/jb.179.5.1431-1441.1997.
[2] Lash LH. Glutathione-Dependent Bioactivation. Curr Protoc Toxicol 2007; 34 (1). https://doi.org/10.1002/0471140856.tx0612s34.

II. PHARMACEUTICAL TECHNOLOGY

II-1

Machine learning-driven optimization of liposomal drug development in microfluidic systems

R. Eugster[1], M. Orsi[1], G. Buttitta[3], N. Serafini[2], M. Tiboni[2], L. Casettari[2], J. L. Reymond[1], S. Aleandri[1], P. Luciani[1]

[1] Department of Chemistry, Biochemistry and Pharmaceutical Sciences, University of Bern, 3012, Bern
[2] Department of Biomolecular Sciences, University of Urbino Carlo Bo, 61029, Urbino, Italy
[3] Department of Chemistry and Technologies of Drugs, Sapienza University of Rome, 00185, Rom, Italy

Introduction: Drug delivery systems efficiently and safely administer therapeutic agents to specific body sites. Liposomes, spherical vesicles made of phospholipid bilayers, have become a powerful tool in this field, especially with the rise of microfluidic manufacturing during the COVID-19 pandemic. However, despite its efficiency, microfluidic liposomal production poses challenges, often requiring laborious optimization on a case-by-case basis. This is due to a lack of comprehensive understanding and robust methodologies, compounded by limited data on microfluidic production with varying lipids. Artificial intelligence (AI) offers promise in predicting lipid behaviour during microfluidic production, with the still unexploited potential of streamlining development.
Aims: Understanding the complex interactions between formulation- and process-parameters, during microfluidic production. Leading to the development of a robust, scalable, and standardized process for microfluidic liposome production, widely adoptable in the pharmaceutical sciences, to accelerate the delivery of therapeutics to clinics and patients.
Methods: Various phospholipids and other excipients, dissolved in ethanol, were mixed with aqueous buffers on diverse microfluidic chips, varying in geometry and manufacturing. Process and formulation parameters were manipulated, and liposomes were characterized using dynamic light scattering (DLS). Herein we employ machine learning (ML) to predict critical quality attributes and process parameters for microfluidic-based liposome production. The principles of explainable AI were applied to interpret the models and understand the underlying mechanisms of liposome formation during microfluidic production.
Results: A comprehensive screening of lipid formulations using microfluidic production revealed trends and patterns among formulation and process parameters, influencing liposomal formation. This in-house dataset was used to train and test ML models. Validated models predicted liposome formation, size, and production parameters, significantly advancing our understanding of lipid behaviour. Extensive model analysis enhanced interpretability and investigated underlying mechanisms, identifying key formulation- and process-parameters as well as molecular features driving the determination of liposome formation and size, among others. Further, a model was established to predict optimal process parameters for desired liposome formulations, supporting the transition from traditional production methods to more scalable and sustainable microfluidic production.
Conclusions: We demonstrated that the integration of predictive modelling with wet lab validation underscores the potential of ML for precise and efficient optimization of microfluidic liposome production, capable of enhancing the development effort, scalability and transitioning to microfluidic technology in pharmaceutical sciences. By leveraging AI, researchers can gain deeper insights into the complex interactions between process parameters and liposome characteristics. The advancements in ML-driven microfluidic production hold promise to accelerate the development of drug delivery systems, making them more accessible and adaptable to a wide range of pharmaceutical applications.

Keywords: Artificial intelligence, machine learning, drug delivery and development, liposomes

Reference:
Eugster R et. al. Leveraging machine learning to streamline the development of liposomal drug delivery systems. bioRxiv 04-Jul-2024. doi: https://doi.org/10.1101/2024.07.01.600773

P-II-2

Intracellular localization of a protein-based DNA delivery system

Ch. Greitens, S. Honrath, M. Burger, J.-C. Leroux

ETH Zürich, Drug Formulation & Delivery, 8093 Zürich

Introduction: Compared to viruses, non-viral gene delivery systems have a higher loading capacity and are considered safer. However, limited knowledge about their intracellular trafficking hampers their progress towards achieving higher efficiency [1]. In our group, a protein-based transfection agent based on the DNA-binding protein Mitochondrial Transcription Factor A (TFAM) was recently introduced. TFAM forms nanoparticles of ~100 nm in diameter when complexed with DNA. It can be fused to the functional proteins phospholipase C (PLC) and vaccinia-related kinase (VRK1) to produce an efficient transfection agent, named "TFAMoplex" [2]. The PLC enabled endosomal escape while the VRK1 mechanism of action which was initially thought to inhibit the intracellular barrier-to-autointegration factor (BAF), remains to be elucidated. The endogenous protein BAF is known to cluster transfected DNA immediately after cytosolic delivery, possibly hindering direct nuclear uptake [3].
Aims and Methods: To understand the DNA delivery process at the cellular level, we studied the association of the TFAMoplex and control Lipofectamine to HeLa cells by flow cytometry and confocal microscopy using Cy3-DNA. Further, we made use of a cell line, stably expressing EGFP-BAF to study the fate of the DNA inside the cytoplasm [4].
Results: The association of Cy3-DNA with the cells was higher for TFAMoplex than for Lipofectamine whereas cells associated with Lipofectamine showed more than 3x brighter mean fluorescence intensity. To differentiate between intracellular and extracellular localization of the transfected DNA, Cy3-DNA that remained on the cellular membrane was counter-stained with an anti-Cy3 antibody and imaged on a confocal microscope. Colocalization analysis revealed that the largest fraction of DNA was localized on the cell surface for both transfection systems after 0.5 and 2 h. On the intracellular level, colocalization analysis of EGFP-BAF clusters with labeled DNA confirmed, that all BAF clusters contain the transfected DNA. Time-lapse imaging showed that cluster formation started approx. 1 h after transfection. With a mScarlet-labeled VRK1, we demonstrated that the protein colocalizes with the BAF cluster. This indicates that the transfected DNA is accompanied by the TFAM fusion proteins even after BAF cluster formation, but also reveals that the VRK1 is incapable to protect the DNA from BAF-mediated clustering.
Conclusions: In summary, we could characterize the transfection process of the TFAMoplex by differentiating the intracellular from extracellular signal of labeled DNA and showed that TFAM proteins are still associated with the DNA inside the cytosol. Further engineering of a VRK1 that remains active in the cytosol might boost transfection efficiency of the protein-based transfection agent.

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Keywords: Non-viral gene delivery, protein-based transfection, intracellular trafficking

References:

[1] Greitens C, Leroux J-C, Burger M. The intracellular visualization of exogenous DNA in fluorescence microscopy. Drug Deliv Transl Res 2023; https://doi.org/10.1007/s13346-024-01563-4
[2] Burger M, Kaelin S, Leroux J-C. The TFAMoplex—Conversion of the Mitochondrial Transcription Factor A into a DNA Transfection Agent. Adv Sci 2022; https://doi.org/10.1002/advs.202104987
[3] Kobayashi S, Koujin T, Kojidani T, Osakada H, Mori C, Hiraoka Y, Haraguchi T. BAF is a cytosolic DNA sensor that leads to exogenous DNA avoiding autophagy. Proc Natl Acad Sci USA 2015; https://doi.org/10.1073/pnas.1501235112

[4] Samwer M, Schneider MWG, Hoefler R, Schmalhorst PS, Jude JG, Zuber J, Gerlich DW. DNA Cross-Bridging Shapes a Single Nucleus from a Set of Mitotic Chromosomes. Cell 2017; https://doi.org/10.1016/j.cell.2017.07.038

P-II-3

Characterization of a biodegradable suction patch for transbuccal peptide delivery

H. Krupke, L. Rabut, D. Klein-Cerrejon, J.-C. Leroux

Department of Chemistry and Applied Biosciences, ETH Zürich, 8093 Zürich

Introduction: Macromolecular drugs, such as peptides, have significantly advanced the treatment of numerous diseases. However, despite their high therapeutic potential, challenges such as low oral bioavailability and substantial variability between and within patients limit their administration to mostly injectable formulations. As injections are often associated with low treatment acceptance, high healthcare costs, and waste production, novel approaches to efficiently deliver these drugs to patients are needed. Recently, we introduced a 3D-printed suction patch designed to enhance the transbuccal diffusion of peptides, offering a potential platform technology to deliver these macromolecular drugs systemically [1].
Aims: The focus of this project was to produce suction patches from biodegradable material to minimize waste. We introduced a highly scalable mold casting method enabling rapid production of buccal patches. To prove the functionality of the novel patches, we investigated their properties with the original prototype as a point of reference.
Methods: The devices were manufactured by mold casting using methacrylated random copolymers of DL-lactide and ε-caprolactone (poly(DLLA-*co*-CL)), which were thermally crosslinked to form polymer networks [2]. Tensile testing was applied on molded test strips to deduct the corresponding mechanical properties. The generated negative pressure, adhesion strength, and biodegradability were tested on molded buccal patches. Loading the patches and applying them *ex vivo* on freshly excised porcine buccal mucosa allowed for analyzing the diffusion depth of the fluorescent drug surrogate cyanine5 formulated with the permeation enhancer sodium taurocholate. Additionally, histochemical staining on buccal tissue sections was employed to quantify the extent of mucosal stretching caused by the patch application.
Results: Thermally crosslinked poly(DLLA-*co*-CL) exhibited high elasticity with elongations at break of up to 250% and Young's moduli of around 2 MPa, thereby mechanically resembling the 3D printed polymer used for the initial prototyping of buccal patches [1]. The devices generated a negative pressure of 40-50 kPa while exhibiting adhesion forces from 1-2 N on a soft silicone surface. Incubation in phosphate-buffered saline revealed a weight loss of around 10% after only two weeks. Most interestingly, *ex vivo* experiments showed drug surrogate diffusion into the buccal epithelial layer and an increase of the tissue surface area by at least 250% across all tested materials.
Conclusions: Our biodegradable suction device shows comparable functionality to the 3D printed prototype. Additionally, its scalable manufacturing process and use of biodegradable materials represent a significant advancement toward more sustainable drug delivery.

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Keywords: Biodegradable, peptide delivery, buccal administration, permeation enhancement

References:

[1] Luo Z, Klein Cerrejon D, Römer S, Zoratto N, Leroux J-C. Boosting systemic absorption of peptides with a bioinspired buccal-stretching device. Sci Transl Med 2023; 15: eabq1887.
[2] Paunović N, Leroux J-C, Bao Y. 3D printed elastomers with Sylgard-184-like mechanical properties and tuneable degradability. Polym Chem 2022; 13: 2271–2276.

P-II-4

Proteomics-supported improvement of TFAMoplex-mediated transfection

S. Honrath, M. Burger, J.-C. Leroux

ETH Zurich, Department of Chemistry and Applied Biosciences, Institute of Pharmaceutical Sciences, Vladimir-Prelog-Weg 3, 8093 Zurich

Introduction: Current non-viral gene delivery systems, including lipid and polymer-based methods, primarily focus on protection, uptake and endosomal escape for DNA delivery. However, these approaches often encounter challenges with efficient cytoplasmic transport and nuclear entry. Our research aims to address these limitations by developing protein-based transfection systems capable of helping DNA throughout the entire delivery process. We introduced the TFAMoplex, a transfection agent derived from mitochondrial transcription factor A (TFAM), which forms approximately 100 nm DNA nanoparticles [1]. The original TFAMoplex featured a bacterial phospholipase for endosomal escape and vaccinia-related kinase 1 (VRK1) to enhance transfection efficiency. However, the precise role of VRK1 in this process remains unclear.
Aims: This study seeks to characterize and enhance the TFAMoplex by replacing VRK1 with dynein light chain proteins, specifically RP3, to improve cytosolic transport by directly tethering the complexes to the dynein motor complex.
Methods: Various TFAMoplex formulations were prepared and characterized for size, zeta potential, and transfection efficiency. Binding kinetics between TFAM-RP3 and dynein intermediate chains were assessed using a luminescence-based assay. Additionally, a proteomics-based assay was implemented to compare protein interactions among different TFAMoplex variants [2].
Results: Significant differences were observed in the transfection efficiencies of TFAMoplexes incorporating different dynein light chain proteins. RP3 was identified as the most effective candidate, with confirmed binding to dynein intermediate chains through both luminescence and proteomics assays. The proteomics analysis also highlighted differences in protein interactors, particularly nucleolar proteins, between the VRK1-containing TFAMoplex and other variants. Incorporating the nucleolar protein leucine-rich repeat-containing protein 59 (LRRC59) into the RP3-TFAMoplex significantly improved transfection efficiency, achieving performance levels comparable to the VRK1-containing system.
Conclusions: Our study shows that TFAMoplexes can be functionally optimized by incorpora-ting alternative protein domains. Direct binding to dynein proteins enhances transfection rates, and the inclusion of the nucleolar protein LRRC59 further boosts transfection efficiency. Moreover, TFAMoplexes outperform commercial transfection agents, such as Lipofectamine.

This project has received funding from the European Research Council (ERC, grant agreement No 884505).

Keywords: Nonviral gene delivery, gene therapy, protein engineering, proteomics

References:

[1] Burger M, Kaelin S, Leroux J-C. The TFAMoplex-Conversion of the Mitochondrial Transcription Factor A into a DNA Transfection Agent. Adv Sci 2022; 9: 1-15
[2] Honrath S, Scherer D, Burger M, Leroux JC. Interaction proteomics analysis to provide insight into TFAMoplex-mediated transfection. J Control Release 2024; 373: 252–264

P-II-5

Developing a novel solubility measurement technique based on second harmonic generation

J. Kalasová[1], O. Tarun[2], M. Kuentz[1]

[1] University of Applied Sciences and Arts Northwest. Switzerland, Institute for Pharma Technology, 4132 Muttenz
[2] Oryl Photonics, 1004 Lausanne

Introduction: Understanding the molecular behaviour and solubility of poorly water-soluble drugs (PWSDs) is crucial as it directly affects their absorption and bioavailability. Traditional solubility measurement methods are time-consuming and require larger amounts of compounds and solvents. We present a novel technique utilizing second harmonic generation (SHG), a nonlinear optical method [1], to measure solvent redistribution around drug molecules, thereby detecting aggregation onset and stable aggregate formation.
Aims: This pilot study aims to: (i) evaluate the efficacy of SHG in determining solubility thresholds of model PWSDs, and (ii) compare SHG results with conventional solubility data.
Methods: Molecular aggregation and solubility of four PWSDs (amiodarone, felodipine, meclizine, miconazole) were measured in phosphate buffer of pH 6.5 at room temperature. Samples, prepared by antisolvent addition method in 96 well-plates, were illuminated with an ultrafast laser (1030 nm, 66.667 kHz, ~250 fs). Optical filters and lenses selectively collected the second harmonic beam (515 nm), which was directed to a photomultiplier tube (PMT). Detection electronics integrated the light collected by the PMT and converted it to voltage. Second harmonic (SH) intensity values were normalized, followed by linear interpolation and baseline calculation. A three sigma (3σ) limit from the baseline determined the solubility threshold.
Results: Solubility was determined for selected PWSDs by assessing the SH intensity as a function of concentration. The values correlated well with literature data. At low concentrations, intensity values remained steady, indicating a linear response (baseline). Near the solubility limit, drug molecules began to form clusters or aggregates, decreasing the system symmetry, resulting in increased SH intensity (deviation from the baseline). For amiodarone, the SH intensity values showed both a clear solubility threshold and a complex pattern at higher concentrations. This pattern suggested transient structural reorganization within the aggregates and a micelle formation, consistent with the amphiphilic structure of amiodarone and its known critical micelle concentration.
Conclusions: This pilot study introduced a novel optical SHG-based technique to measure the solubility of selected drugs. The measured solubilities aligned well with literature values, demonstrating advantages in precision, speed, and reduced resource consumption over traditional methods. Additionally, the technique offered insights into the molecular dynamics of drug molecules in solution phase, though further research is needed to fully explore its potential.

Keywords: Poorly water-soluble drugs, solubility measurement, second harmonic generation, nonlinear optics, molecular aggregation

Reference:

[1] Lee CJ, Strachan CJ, Manson PJ, Rades T. Characterization of the bulk properties of pharmaceutical solids using nonlinear optics – a review. J Pharm Pharmacol 2010; 59(2): 241-250.

P-II-6

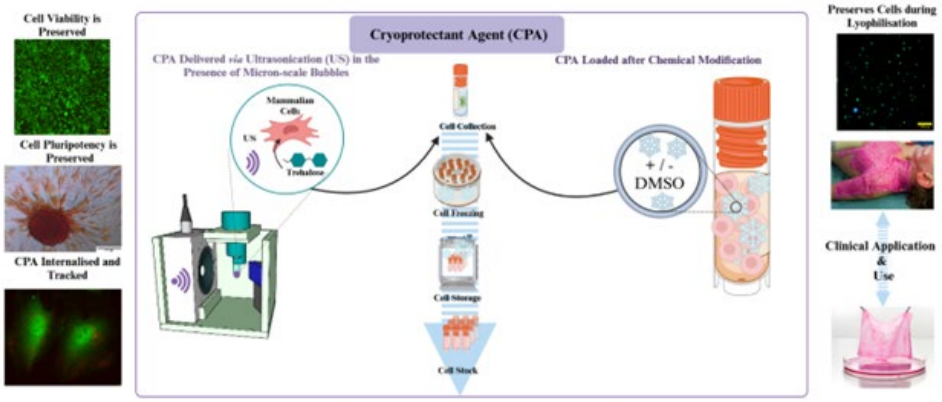
Physics versus chemistry: Innovations in cryopreservation for cell-based therapies via ultrasonication or chemical modification

V. Patrulea[1,2,3], C.V. Fuenteslópez[1], M. Gray[1], M. Flahaut[4], A. Laurent[4,5], C. Scaletta[4], N. Hirt-Burri[4], S. Bahcevanci[1], A. Martin[1,6], C.A.B. Smith[1], C. Coussios[1], Z. Cui[1], H. Ye[1], L.A. Applegate[4,7]

[1] *University of Oxford, Institute of Biomedical Engineering, Department of Engineering Science, OX3 7DQ Oxford, UK*
[2] *University of Geneva, Institute of Pharmaceutical Sciences of Western Switzerland, 1205 Geneva*
[3] *University of Geneva, School of Pharmaceutical Sciences, 1205 Geneva*
[4] *University of Lausanne, Regenerative Therapy Unit, Lausanne University Hospital, 1066 Epalinges*
[5] *Manufacturing Department, TEC-PHARMA SA, 1038 Bercher*
[6] *Institute of Diagnostic and Interventional Radiology, University Hospital Zurich, 8091 Zurich*
[7] *University of Lausanne, Lausanne Burn Center, Lausanne University Hospital, 1011 Lausanne*

Introduction: Advances in cell culture techniques — like isolation, expansion, banking, and transport — are crucial for regenerative medicine. While dimethylsulfoxide (DMSO) is a common cryoprotective agent (CPA), its long-term cytotoxicity can affect cell function. Effective cryopreservation is a key to maintaining cell quality and therapeutic efficacy. As the field evolves, a detailed, cell-specific overview of cryopreservation methods is essential to avoid trial-and-error and maximize the potential of cell-based therapies [1, 2].
Aim: Delivery of CPAs through physical or chemical methods.
Methods: Trehalose, a natural disaccharide, has been proposed as a non-toxic CPA, but the lack of specific cell membrane transporter receptors inhibits transmembrane transport and severely limits its cryoprotective capability. This research presents a novel method for utilising trehalose as a non-toxic CPA and its successful delivery into mesenchymal stem cells (MSCs) using ultrasound (US) in the presence of microbubbles (MB). Another approach was to deliver a safe CPA like CryoOx to MSCs, without using DMSO, after chemically modifying it.
Results: The optimized CPA concentration effectively preserved not only membrane integrity and cell viability but also the multipotency of MSCs, which is crucial for stem cell therapy. Additionally, confocal imaging revealed that rhodamine-labeled trehalose was transported into the cells, not merely attached to the cell membrane. We also successfully lyophilized cells and maintained their membrane integrity. Overall, this study demonstrates that chemically modifying CPAs or using physical delivery methods provides promising cryoprotective capabilities without the cytotoxicity associated with DMSO-based methods.

Fig. 1. Overview of the CPA delivery via ultrasonication (US) or after chemical modification.



Conclusions: Our findings show that both chemically modified [1] and ultrasound-induced [2] CPAs are promising alternatives, providing cell viability similar to commercial CPAs. This research highlights the need for secure, efficient cryopreservation techniques in cell banking to enhance quality, ensure patient safety, and advance regenerative medicine.

Keywords: Cell cryopreservation, tissue engineering, long-term storage, cryoprotectant, lyophilisation.

References:

[1] Flahaut et al. J Invest Dermatol 2024; Accepted.
[2] Fuenteslopez et al. Comms Eng (Nature) 2024; Accepted.

P-II-7

Lipid-degrading enzymes as permeation enhancers

I. Bortolazzo, S. Fraefel, M. Bohley, M. Heussi, D. Gao, J.-C. Leroux

Institute of Pharmaceutical Sciences, Department of Chemistry and Applied Biosciences, ETH Zurich, Zürich

Introduction: Therapeutic peptides are highly valuable therapeutics. While peptides are generally dosed parenterally, finding ways to deliver them orally would be an attractive approach to improve patient acceptance and compliance [1]. However, unfavorable physicochemical properties such has high molecular weight and hydrophilicity together with the harsh environment in the gastrointestinal (GI) tract pose significant challenges. Barriers encountered in the GI tract are digestive enzymes and rapid changes in pH, the mucus as a diffusion barrier, and the intestinal epithelium which presents the main obstacle for effective peptide absorption [1]. Enterocytes contain lipid rafts – special membrane regions rich in sphingolipids, glycolipids, and cholesterol - that can further enhance the barrier properties [2]. To date, the most successful approach to overcoming the epithelial barrier is by using chemical permeation enhancers (PE) such as salcaprozate sodium (SNAC), sodium caprate (C10) and sodium caprylate (C8) [2]. PEs transiently open tight junctions and/or disrupt cell membrane integrity and increase membrane fluidity [3]. Despite the use of PEs, the oral bioavailability of commercially available peptide formulations remains around 1% [4]. Therefore, there is a persisting need in finding novel and more efficient PEs. Taking inspiration form nature where various pathogens use a mix of enzymes to disrupt the cell membrane integrity [5, 6], we showcase that the lipid-degrading enzymes Phospholipase C (PLC) from *Bacillus cereus* and Sphingomyelinase C (SMase) from *Staphylococcus aureus* are potent PEs.
Aims: To assess the suitability of using enzymes as intestinal permeation enhancers (PEs), we first evaluated the activity and stability of PLC under GI mimicking conditions. Next, we investigated the potential of SMase alone and in combination with PLC as PEs *in vitro* and *ex vivo*.
Methods: The enzymes were recombinantly produced. PLC activity and stability was investigated under different GI mimicking conditions. The impact of SMase on membrane integrity, alone or in combination with PLC, was tested on Caco-2 cells by assessing the induced lactate dehydrogenase release (LDH assay) and its impact on cell viability. Further, transepithelial-electrical resistance (TEER) and cell permeability of FD4 and FD10 were measured. Additionally, immunostainings were used to determine if the enzymes affected the opening of tight junctions. *Ex vivo* experiments were conducted in the Ussing Chamber, where the TEER reduction of porcine intestinal tissue was assessed with the enzymes separately and in combination.
Results: We confirmed PLC activity under various GI mimicking conditions. SMase alone showed a similar reduction in epithelial integrity of Caco-2 cells compared to PLC, while the combination with PLC resulted in additive effects on membrane integrity. The results of the cell permeability assay revealed a 2-fold increase in permeability for FD4 and FD10 when using the combination of both enzymes. *Ex vivo* studies using freshly excised porcine small intestinal tissue and the Ussing chamber confirmed the observation that a combination of both enzymes is beneficial. Overall, we demonstrated that lipid-degrading enzymes are a promising class of new PEs, with a combination of the two showing promise for further *in vivo* testing.

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Keywords: Oral peptide delivery, permeation enhancers, phospholipase C, sphingo-myelinase C, Ussing Chamber

References:

[1] Brayden D and O'Mahony D. Novel oral drug delivery gateways for biotechnology products: polypeptides and vaccines. PDA JPST 1998; 1(7): 291-299.

[2] Bohley M and Leroux JC. Gastrointestinal permeation enhancers beyond sodium caprate and SNAC-what is coming next?. Adv Sci 2024; Advanced online publication DOI: 10.1002./advs.202400843.

[3] Maher S, Geoghegan C and Brayden DJ. Intestinal permeation enhancers to improve oral bioavailability of macromolecules: reasons for low efficacy in humans. Expert Opin Drug Deliv 2021; 18(2): 273-300.

[4] Twarog C, Fattah S, Heade J et al. Intestinal permeation enhancers for oral delivery of macromolecules: a comparison between salcaprozate sodium (SNAC) and sodium caprate (C10). Pharmaceutics 2019; Advanced online publication DOI: 10.3390/pharmaceutics11020078.

[5] Bock J, Liebisch G, Schweimer J et al. Exogenous sphingomyelinase causes impaired intestinal epithelial barrier function. World J Gastroenterol 2007; 13(39): 5217–5225.

[6] Flores-Díaz M, Monturiol-Gross L, Naylor C et al. Bacterial sphingomyelinases and phospholipases as virulence factors. Microbiol Mol Biol Rev 2016; 80(3): 597–628.

P-II-8

Modelling fibrosis in endometriosis using a 3D cell culture system based on fibronectin (FN)-silk

S. Teworte[1], M. C. Behrens[1], M. Widhe[2], M. Hedhammar[2], P. Luciani[1]

[1] Department of Chemistry, Biochemistry and Pharmaceutical Sciences, University of Bern, 3012 Bern
[2] Department of Protein Science, School of Engineering Sciences in Chemistry, Biotechnology and Health, KTH Royal Institute of Technology, AlbaNova University Center, SE-114 21 Stockholm, Sweden

Introduction: Endometriosis is a gynaecological disease affecting 10% of women of reproductive age, characterized by fibrotic lesions at ectopic sites in the pelvis. Fibrosis is increasingly recognised as a potential target for pharmacological therapies in endometriosis. We hypothesized that 3D cell models of endometriosis could better recapitulate the fibrotic phenotype of the disease than traditional cell monolayers, thus enhancing the preclinical evaluation of antifibrotic drug candidates.
Aims: Our aim was to explore two 3D cell culture formats to model fibrotic pathophysiology in endometriotic stroma and epithelium. In the first format, cells were integrated into a network of the recombinantly produced spider silk protein FN-silk, genetically functionalized to harbour the Arg-Gly-Asp (RGD) cell binding motif from fibronectin. The second format was a spheroid. Additionally, we aimed to evaluate the utility of these 3D cell models in assessing pirfenidone, an antifibrotic drug candidate with the potential to be repurposed for endometriosis treatment.
Methods: We developed FN-silk networks and spheroids with endometrial stromal and endometriotic epithelial cell lines, characterizing them in terms of morphology and metabolic activity. We examined cellular processes involved in fibrotic pathophysiology – including fibroblast-to-myofibroblast activation, extracellular matrix protein expression, epithelial-to-mesenchymal transition, and intracellular signalling – at the RNA and protein levels by RT-qPCR and immunofluorescent staining. This allowed us to compare different culture formats regarding their fibrotic phenotype and response to pirfenidone treatment.
Results: FN-silk networks integrated 90-100% of the cells seeded and supported cell proliferation in 3D, whereas cells in spheroids exhibited a decline in metabolic activity over 7 days. Culture in 3D upregulated fibrotic markers at the transcriptomic level in both stromal and epithelial cells. For stromal cells, fibrosis was induced in FN-silk networks by TGF-β1 treatment, while cells in spheroids did not respond. For epithelial cells, TGF-β1 treatment induced fibrosis similarly in both FN-silk networks and spheroids. Pirfenidone demonstrated antifibrotic effects in cells in FN-silk networks, suggesting its suitability as a potential therapy to reverse fibrosis in endometriosis.
Conclusions: Cells cultured in FN-silk networks are superior to spheroids in modelling the fibrotic phenotype of endometriotic stroma and epithelium *in vitro*. This work paves the way for more complex *in vitro* models of endometriosis, allowing us to begin to model fibrotic lesion architecture. Additionally, cells in FN-silk networks may have translational potential to model fibrotic pathologies of other organs.

Keywords: Fibrosis, endometriosis, 3D cell culture, silk protein, spheroid, pirfenidone

P-II-9

Heat application for enhanced oral peptide absorption – a proof-of-concept

D. Gao, S. Fardel, M. Bohley, J.-C. Leroux

Department of Chemistry and Applied Biosciences, ETH Zurich, 8093 Zurich

Introduction: Macromolecular therapeutics, such as peptide and protein drugs, play a crucial role in the treatments of a broad range of diseases. Despite their high therapeutic potential and specificity, their administration is almost exclusively limited to injections, as oral delivery is strongly restricted by gastrointestinal barriers presented by digestion, mucus, and epithelial linings [1]. To enhance permeation over the epithelial barrier, we propose heat application as novel strategy to enhance oral delivery of macromolecular therapeutics.
Aims: We propose heat application as novel permeation enhancing mode to boost macromolecular diffusivity. This study aims at to demonstrate the efficient reduction of epithelial integrity and permeation of macromolecules under heat application. Furthermore, this work will elucidate the permeation enhancement mechanism of heat application and investigate synergistic effects with commercial permeation enhancers.
Methods: Application of heat was first studied on a monolayer of colorectal adenocarcinoma (Caco-2) cells cultured on Transwell inserts using a custom build heating setup. Tested heat conditions include 39 °C and 42 °C and heating duration of 2 h. The transepithelial resistance (TEER) was measured during heating to monitor changes of epithelial integrity. Permeability assays with Cy5-labelled polyethylenglycols (PEGs) with different molecular weights (1 kDa, 5 kDa, and 20 kDa) as surrogates for macromolecular drugs were performed to assess the efficacy of heat as permeation enhancement method. Additionally, heat was combined with commercial permeation enhancers to detect possible synergies of combing different strategies. Finally, mechanistic studies were performed to investigate the influence of heat on membrane integrity, tight junction proteins and their expression.
Results: Cells heated for 2 h at 39 °C and 42 °C showed a significant decrease in TEER of up to 34% and 66% respectively. Combining heat with C10 as a chemical permeation enhancer showed that the decrease in TEER caused by 10mM C10 could be reached with only 2.5 mM and at 39 °C. Subsequent permeation assays with Cy5-PEGs with molecular weight up to 20 kDa revealed permeation enhancement of up to a factor of 3 over 2 h at 42 °C. After heat application, cell monolayer showed full recovery after 24 h. Additionally, cytotoxicity assays revealed that permeation enhancement was not linked to cell death.
Conclusion: We found that modest heat applications of 42 °C for 2 h could significantly reduce the barrier properties of the epithelium in vitro and increases the macromolecular permeation. We thereby propose heat as a novel physical mode to improve oral peptide absorption in the gastrointestinal tract.

Keywords: Oral delivery, physical mode, permeation enhancer

Reference:
[1] Drucker DJ. Nat Rev Drug Discovery 2020; 19(4): 277–289.

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P-II-10

3D-printed lipid mesophases for the treatment of chronic liver disease

R. Gazzi[1], M. Carone[1], R. Eugster[1], R. Gelli[2], N. Manten[1], A.A. Ganguin[1], S. Di Valerio[3], G. Yadav[1], P. Castaldo[4], R. Mezzenga[5], P. Luciani[1], S. Aleandri[1]

[1] Department of Chemistry, Biochemistry and Pharmacy, University of Bern, 3012 Bern
[2] Department of Chemistry Ugo Schiff and CSGI, University of Florence, 50019 Florence, Italy
[3] Department of Clinical and Molecular Sciences, University «Politecnica delle Marche», 60126 Ancona, Italy
[4] Department of Biomedical Sciences and Public Health, University «Politecnica delle Marche», 60126 Ancona, Italy
[5] Laboratory of Food & Soft Materials, ETHZ, 8092 Zurich

Introduction: Lipid-based formulations offer a promising strategy for improving the oral bioavailability of lipophilic drugs, but their integration into solid oral dosage forms faces challenges due to high viscosity and heat sensitivity. This study employed semi-solid extrusion to produce 3D-printed tablets, or printlets, rich in bioactive lipids [1]. A lipid mesophase (LMP)-based ink of S80, vitamin E, and water, was used for additive manufacturing. In the context of chronic liver disease treatment, the inclusion of S80, a natural phospholipid, is interesting due to the deactivation of profibrogenic hepatic stellate cells [2]. The printlets' efficacy in delivering poorly water-soluble drugs through a self-emulsification process was exemplified by the incorporation of obeticholic acid.
Aims: To develop an LMP-based solid oral dosage form using semisolid extrusion 3D-printing for the treatment of chronic liver disease, capable of enhancing drug solubility and bioavailability through a self-emulsification process.
Methods: The LMP composition was screened to optimize printability, shape retention, and disintegration. The formulation was characterized in terms of drug content and homogeneity, by HPLC, phase identity by SAXS, and rheological properties. The printlets' behavior in the gastrointestinal tract and the self-emulsification mechanism were evaluated via SAXS, Cryo-TEM, and drug release profiling in simulated fluids. *In vitro* tests assessed effects on intestinal barrier integrity and hepatic cell antifibrotic activity.
Results: A ternary system consisting of 68% S80, 20% water, and 12% vitamin E exhibited optimal mechanical properties. Homogeneous drug distribution within the printlet was confirmed. SAXS measurements revealed the coexistence of inverse hexagonal and lamellar phases, crucial for printability and disintegration. Rheological characterization demonstrated that the printing process enhanced the structural strength of the LMP. Printlets remained intact in acidic conditions with no drug release in simulated gastric fluid, but fully disintegrated in simulated intestinal fluid within 6 h. The self-emulsification process was confirmed by the abundance of micelles and vesicles in Cryo-TEM images. This colloidal system did not compromise the viability and permeability of intestinal epithelial cells. Moreover, antifibrotic activity in LX-2 cells was observed, as evidenced by increased expression of the PLIN2 gene and the presence of cytoplasmic lipid droplets.
Conclusions: This study effectively applied lipid mesophases for semi-solid extrusion 3D-printing, showing promise in delivering poorly water-soluble drugs through self-emulsification. With enhanced drug solubility, coupled with co-formulation with hepatoprotectants, the printlets hold potential as an oral treatment for chronic liver disease.

Keywords: Lipid mesophases, 3D-printing, semi-solid extrusion, oral delivery, chronic liver disease.

References:

[1] Carone M, Gazzi R, Eugster R, Gelli R, Manten N, Ganguin A, Di Valerio S, Yadav G, Castaldo P, Mezzenga R, Luciani P, Aleandri S. Adv MatTechnol 2024. Doi: doi.org/10.1002/admt.202301930.
[2] Valentino G, Zivko C, Weber F, Brülisauer L, Luciani P. Pharmaceutics 2019; 11: 676.

III. PHARMACOEPIDEMIOLOGY

P-III-1

Market uptake and pattern of use of emerging topical rosacea medications in Switzerland: A descriptive study using Swiss claims data

T. Zappalà[1], A. A. Navarini[2], C. A. Huber[3], C. Meier[1,4], J. Spoendlin[1,4]

[1] *Basel Pharmacoepidemiology Unit, Division of Clinical Pharmacy and Epidemiology, Department of Pharmaceutical Sciences, University of Basel, Basel*
[2] *Department of Dermatology, University Hospital Basel, Basel*
[3] *Department of Health Sciences, Helsana Group, Zurich*
[4] *Hospital Pharmacy, University Hospital Basel, Basel*

Introduction: Rosacea is a chronic inflammatory facial skin and ocular disease. In Switzerland, topical metronidazole was the only first-line topical treatment for rosacea, until topical brimonidine was licensed in 2014 and topical ivermectin in 2016.
Aims: To evaluate the use of topical medications specifically indicated for rosacea between 2015 and 2021 in Switzerland.
Methods: We used claims data from the Swiss health insurance 'Helsana' between January 2015 and December 2021 (study period). We quantified the period prevalence of treated rosacea in Switzerland, overall and stratified by age (≥/<50 y) and sex. Therefore, we identified all persons with at least one claim of either topical metronidazole, brimonidine, or ivermectin, divided by all continuously enrolled persons during the study period. We also quantified the yearly prevalence of treated rosacea among persons insured during the entire calendar year. We quantified the proportion of patients with monotherapy or with combinations of these rosacea treatments, as well as the proportion of rosacea patients with claims for topical corticosteroids within the same calendar year.
Results: The period prevalence of treated rosacea was 3.0% (22,470 / 749,598) between 2015 and 2021. Of all rosacea patients, 70% were female and 71% were aged ≥50 years. The yearly prevalence of treated rosacea increased from 0.52% in 2015 to 0.62% in 2021. Metronidazole was used by 95% and 93% of all treated rosacea patients in 2015 and 2016 and then continuously dropped to 63% in 2021. Ivermectin was claimed by 24% in 2017 and increased to 41% in 2021. The proportion of topical monotherapy for rosacea did not change much during the study period with 92% in 2015 and 88% in 2021. However, while metronidazole monotherapy accounted for 92% of all rosacea patients, it decreased to 55% in 2021, whereas ivermectin monotherapy accounted for 33%. Only between 4% and 6% of all rosacea patients combined metronidazole and ivermectin in the same year. Brimonidine was claimed by <10% of all treated rosacea patients mainly as monotherapy throughout. Topical corticosteroids were claimed by 31% of all treated rosacea patients in 2015, which did not change during the study period (29% in 2021).
Conclusion: The discrepancy between yearly and period prevalence suggests that many rosacea patients receive intermittent therapy. Despite the decline in use, metronidazole monotherapy remains the most frequently used topical rosacea treatment in Switzerland. However, the use of ivermectin is continuously increasing, while brimonidine accounts for a small and stable proportion of use. Although contraindicated for rosacea, almost one third of rosacea patients also claimed topical corticosteroids in the same year, but the indication for the steroid treatment is unknown.

Keywords: Rosacea, drug utilization study, healthcare claims data

P-III-2

Development of a real-time opioid monitoring system at a Swiss cantonal hospital: An observational study

M. Rainer[1,2], L. Gasparovic[1], L. Gurtner[1,2], D. Stämpfli[1,2], A.M. Burden[1]

[1] Institute of Pharmaceutical Sciences, ETH Zurich, 8093 Zurich
[2] Hospital Pharmacy, Department of Medical Services, Kantonsspital Baden AG, 5404 Baden

Introduction: Overprescribing of opioid analgesics can cause significant population-level harm, as illustrated by the opioid crisis in North America. Hospitalization is often an entry point for new or higher opioid prescriptions, for example in the context of a medical emergency or surgery. Therefore, opioid stewardship programs have become critical to curb the rising opioid use. However, in Switzerland, there are no established opioid stewardship programs, and little is known about the use of opioids during hospitalization.
Aims: We implemented an opioid dashboard at the Kantonsspital Baden (KSB) to assess opioid consumption in order to monitor in-hospital opioid use, describe patient demographics, and identify prescribing hotspots within the KSB.
Methods: We extracted all patient records with an administration of opioid analgesics between 1 January 2023 and 31 December 2023. All opioid administrations are captured in the clinical information system, including the substance, dose, and route of administration, prescriber information (e.g. medical specialty), and patient-level information (e.g. age, sex, ward). We descriptively analyzed the patient characteristics for all administrations and identified the medical specialties with highest prescribing. We subsequently examined patients with three or more consecutive days of opioid use, as these represent a viable population for opioid stewardship.
Results: A total of 2,401 patients (60% female) were identified. The median patient age was 69 years (IQR 55-79). The largest proportion of patients with opioids were found in the emergency department (58.8%, n=1,411), followed by orthopedics (7.7%, n=184), and general surgery (7.6%, n=182). The most frequently administered substances were oxycodone + naloxone (33.2%), morphine (19.9%), and buprenorphine (16.4%). Patients received opioids for a mean of two consecutive days (SD 1.4 days). A total of 608 patients (65% female) with three or more consecutive days of opioid use were identified with a median age of 71 years (IQR 59-80). This population most frequently presented with unspecified spondylopathies, back pain, and arthrosis of the knee.
Conclusions: Using a novel opioid dashboard, we identified over 2,000 patients with opioids administered at the KSB in 2023, of which over 25% received opioids for three or more days, highlighting the potential need for opioid stewardship programs. We could also identify patient populations and wards with high opioid use, allowing the development of targeted interventions to reduce opioid burden.

Keywords: Opioid stewardship, drug safety, clinical pharmacy, pharmacoepidemiology, drug monitoring, opioid analgesics

P-III-3

Uptake of pertussis vaccine during pregnancy following guideline changes in Switzerland: A descriptive study using Swiss claims data

N.A. Stohler[1], S. Graber[2], C.A. Huber[2], D. Surbek[3], U. Heininger[4], C.R. Meier[1,5,6], J. Spoendlin[1,5]

[1] *Basel Pharmacoepidemiology Unit, Division of Clinical Pharmacy and Epidemiology, Department of Pharmaceutical Sciences, University of Basel, 4031 Basel*
[2] *Department of Health Sciences, Helsana Group, 8081 Zürich*
[3] *Department of Obstetrics and Gynaecology, Bern University Hospital, Insel Hospital, University of Bern, 3010 Bern*
[4] *University of Basel Children's Hospital, Paediatric Infectious Diseases and Vaccinology, and University of Basel Medical Faculty, 4056 Basel*
[5] *Hospital Pharmacy, University Hospital Basel, 4031 Basel*
[6] *Boston Collaborative Drug Surveillance Program, MA 11 Muzzey St, Lexington, MA 02421, USA*

Introduction: Pertussis infections in newborns are associated with a greater risk for complications and may be life-threatening. Since 2017, the Swiss NITAG guidelines and the Swiss Society of Obstetrics and Gynecology (SGGG) therefore recommend a dose of pertussis vaccine in the late second or early third trimester in every pregnancy to protect the newborn from infection during the first weeks of life. Prior to this, the 2013 guidelines recommended vaccination if the last dose was more than five years ago.
Aims: We aimed to evaluate the coverage of pertussis vaccines during pregnancy in Switzerland between 2012 and 2022.
Methods: We conducted a descriptive, retrospective study using anonymized healthcare claims data from one of the largest health insurance companies in Switzerland, the Helsana Group (2012-2022). We established a cohort of pregnancies by identifying deliveries (live and stillbirths, not including abortions) and estimating the date of the last menstrual period (LMP; for births after the start of 2020, the true LMP date was recorded). Pertussis vaccines were captured based on ATC codes recorded during pregnancy, and exposure prevalence was quantified as the number of vaccinated pregnant women out of all pregnancies. We quantified exposure prevalence to pertussis vaccines overall during pregnancy, as well as stratified by pregnancy week, calendar year, canton of residence, and maternal age.
Results: We included a total of 113,368 pregnancies between 2012 and 2022. During the study period, 30.7% of pregnant women had a pertussis vaccine recorded during pregnancy. The median time point of pertussis vaccine administration was gestational week 28 (IQR 25-32). In 2012, the exposure prevalence to pertussis vaccines was 0.6%, which increased to 15.1% in 2017. After the 2017 guideline changes, vaccination prevalence continuously increased to 56.1% in 2020, after which it plateaued. Cantons in the French-speaking part of Switzerland exhibited the highest proportion of vaccinated pregnancies, with 70.9% in Geneva, 64.6% in Neuchatel, and 72.2% in Vaud among women who gave birth in 2022. Conversely, rural cantons in the German-speaking part of Switzerland, including Appenzell Innerrhoden, Appenzell Ausserrhoden, and Glarus, revealed the lowest vaccination rates around or below 30% among women who gave birth in 2022. Maternal age was not meaningfully associated with the proportion of vaccinated pregnancies.
Conclusion: There has been a significant increase in pertussis vaccinations during pregnancy in Switzerland following the 2017 guideline update. However, vaccination coverage remains uneven across different regions, with notably lower proportions in rural German-speaking cantons. Similar regional disparities have also been shown for childhood and other vaccines and may warrant more targeted public health interventions.

Keywords: Pertussis, pregnancy, vaccination, claims database, observational study

P-III-4

Are specific DOACs favored by clinicians for patients with atrial fibrillation post stroke? A secondary data analysis of the MAAESTRO-study in Basel, Switzerland

K. Rekk[1], E. Dülger[1], A.A. Polymeris[2], P.A. Lyrer[2], S.T. Engelter[2,3], S.S. Allemann[1], I. Arnet[1]

[1] *Department of Pharmaceutical Sciences, Pharmaceutical Care Research Group, University of Basel, 4056 Basel*
[2] *Department of Neurology and Stroke Centre, University Hospital Basel and University of Basel, 4056 Basel*
[3] *Department of Geriatric Medicine, Neurology and Neurorehabilitation, Felix Platter Hospital, University of Basel, 4055 Basel*

Introduction: To lower the risk of a recurrent stroke, patients receive direct oral anticoagulants (DOAC) as part of their secondary prevention therapy. Currently, there are four DOAC agents available on the market in Switzerland: Three factor Xa inhibitors (Apixaban twice daily, Rivaroxaban once daily and Edoxaban once daily) and one factor II inhibitor (Dabigatran twice daily). Latest guidelines do not recommend the use of one specific DOAC agent. MAAESTRO was a RCT that investigated the effect of a pillbox and educational intervention. Patients with atrial fibrillation (AF) hospitalized for a stroke at the University Hospital Basel, Switzerland, were enrolled in the MAAESTRO study from 2018-2022 with focus on adherence to DOAC over one year.
Aims: To analyse prescription preferences for DOAC to AF-patients after a stroke, using the MAAESTRO data.
Methods: We compared the mean values of demographic and clinical variables between the patients according to the prescribed DOAC using the Kruskal-Wallis Test for multiple comparison and Dunn-Bonferroni Post-Hoc Test for pairwise comparison or the Fishers Exact Test.
Results: Data from 84 patients were analysed. During the study, all four DOAC agents were prescribed. Most patients were treated with Apixaban (43; 51%), followed by Dabigatran (19; 23%), Rivaroxaban (12; 14%), and Edoxaban (10; 12%). Dabigatran was prescribed more often to younger patients with better renal function compared to Apixaban (*) and Edoxaban (**) [mean age (70.0±10.5 years *vs.* 78.4±7.3 years *vs.* 79.33±10.0 years; p=0.021*; p=0.045**), and mean eGFR (84.2±11.9 mL/min *vs.* 65.7±19.2mL/min *vs.* 58.4±20.6 mL/min; p=0.001*; p=0.002**)]. Apixaban was prescribed more often to patients with a higher CHA_2DS_2VASc-Score compared to Dabigatran (mean: 5.7±1.3 *vs.* 4.6±1.0; p=0.024). Seventy-four percent of patients were prescribed a twice-daily DOAC (Apixaban and Dabigatran) while 26% received a once-daily DOAC (Rivaroxaban and Edoxaban). We found no differences for the prescription of Rivaroxaban compared to the other agents.
Conclusions: Clinicians showed a preference in the prescription of DOAC as over 50% of the study participants obtained Apixaban. Individual factors like age, renal function and CHA_2DS_2VASc-Score seem to influence the DOAC selection. Clinicians seem to favor twice-daily DOAC compared to once-daily agents.

Keywords: DOAC, prescription, stroke, AF

P-III-5

The importance of assessing drug-drug-gene interactions

N.L. Wittwer[1,2], C.R. Meier[1,2,3], C.A. Huber[4], J. Moser[1], H.E. Meyer zu Schwabedissen[5], S. Allemann[6*], C. Schneider[1,2*]

[1] Basel Pharmacoepidemiology Unit, Division of Clinical Pharmacy and Epidemiology, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel
[2] Hospital Pharmacy, University Hospital Basel, 4031 Basel
[3] Boston Collaborative Drug Surveillance Program, 02421 Lexington, MA
[4] Department of Health Sciences, Helsana Group, 8001 Zürich
[5] Biopharmacy, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel
[6] Pharmaceutical Care, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel
**These authors contributed equally to this work.*

Introduction: Drug-drug-gene interactions may lead to phenoconversion, which can limit the applicability of pharmacogenetic (PGx) testing in clinical practice. However, the prevalence and impact of drug-induced phenoconversion remain largely unclear.
Aims: The aim of this study was to determine the prevalence of interactions between PGx drugs associated with CYP2C9, CYP2C19, and CYP2D6 and drugs that act as inhibitors or inducers of those enzymes in the Swiss population.
Methods: We conducted a retrospective, descriptive study using claims data from the health insurance Helsana between 1 January 2017 and 31 December 2021. The concomitant use of PGx drugs and inhibitors/inducers was defined as instances where a claim of a PGx drug and a claim of an inducer or inhibitor concerning the same enzyme were made within a specified temporal window, either ± 5 days or ± 30 days. Moreover, we examined the specific drugs involved in these interactions and characterised the individuals affected.
Results: A total of 894'748 individuals were continuously insured. Between 17.4% (± 5-days window) and 24.8% (± 30-days) of individuals were exposed to interacting drug pairs. Among those, 1.5% to 2.2% were exposed to strong interacting drug pairs over the course of the five-year period. Individuals exposed to interacting drugs were more frequently female (62.0 to 62.1% *vs.* 52.4%), older (57.8 ± 21.1 to 58.7 ± 21.1 *vs.* 44.5 ± 24.0 years) and took more drugs (37.0 ± 18.1 to 38.6 ± 18.9 *vs.* 19.7 ± 16.7 drugs) than the general population. The interactions were more frequently associated with CYP2D6 or CYP2C19 than with CYP2C9. The drug pairs most frequently involved in interactions were pantoprazole-quetiapine, citalopram-quetiapine, and metoprolol-quetiapine within the ± 30-days window, or pantoprazole-quetiapine, escitalopram-quetiapine, and citalopram-quetiapine within the ± 5-days window.
Conclusions: The high prevalence of the simultaneous use of PGx drugs with inhibitor and inducer drugs demonstrates the necessity of considering non-genetic factors, such as drug-induced phenoconversions, when interpreting PGx test results.

Keywords: Pharmacogenetic, drug-drug interaction, drug-drug-gene interaction, phenoconversion

P-III-6

Antidepressant drug switching in the Swiss population with a focus on Escitalopram and pharmacogenetically relevant drugs: A drug utilization study using the Helsana Database

M. Roth[1,2], N.L Wittwer[1,2], C.R. Meier [1,2,3], C.A. Huber[4], H.E. Meyer zu Schwabedissen[5], M.L. Lampert[6], S. Allemann[6*], C. Schneider[1,2*]

[1] Basel Pharmacoepidemiology Unit, Division of Clinical Pharmacy and Epidemiology, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
[2] Hospital Pharmacy, University Hospital Basel, 4056 Basel
[3] Boston Collaborative Drug Surveillance Program, 02421 Lexington, MA, USA
[4] Department of Health Sciences, Helsana Insurance Group, 8001 Zürich
[5] Biopharmacy, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
[6] Pharmaceutical Care, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
**These authors contributed equally to this work*

Introduction: Depression affects approximately 20% of the Swiss population [1]. Selective serotonin reuptake inhibitors (SSRIs) are recommended as first-line treatments. However, only 30-40% of patients taking SSRIs achieve remission [2, 3]. Pharmacogenetic variation may explain the high rate of treatment failure for pharmacogenetically relevant (PGx) SSRIs such as escitalopram.
Aims: The aim of this study was to assess how many people in Switzerland switch from escitalopram to other antidepressants, and to assess whether there are any differences between PGx and non-PGx antidepressants.
Methods: We identified all persons who were registered with Helsana, a Swiss health insurance company, between 1 July 2017 to 30 June 2019, and who had at least one outpatient drug claim for escitalopram and at least one other antidepressant during the study period. All antidepressants were classified as PGx relevant or not according to information provided by PharmGKB. We used descriptive statistics to quantify the number of patients switching from escitalopram to other antidepressants.
Results: We identified 15'275 persons (66.7% women) with a mean age of 57.1 (± 20.4) years taking escitalopram in addition to at least one other antidepressant during the study period. In total, they claimed 182'607 antidepressant drug prescriptions during the study period, of which 102'490 (56.1%) were for PGx relevant antidepressants, and 71'858 (39.4%) for escitalopram. The mean number of antidepressant prescriptions claimed per person was 5.0 (± 12.5). During the study period, 4774 (31.3%) persons switched from escitalopram to another antidepressant, 14.3% to another SSRI, 8.5% to a NSRI, and 77.2% to other antidepressants. In total, 1241 persons switched from escitalopram to other PGx-relevant antidepressants, 41.7% switched to a PGx-SSRI, 32.8% to a PGx-NSRI, and 25.5% to another PGx-antidepressant. On the other hand, 3533 switched to non-PGx-relevant antidepressants, but only 4.7% switched to a non-PGx-SSRI or -NSRI, the rest switched to another non-PGx antidepressant.
Conclusions: The occurrence of drug switches from escitalopram to other antidepressants in Switzerland is high. In 74% of the switches, the patient switches to an antidepressant that is not pharmacogenetically relevant.

Keywords: Pharmacogenetics, drug switches, antidepressants, escitalopram, Switzerland, claims data, Helsana

References:
[1] Schuler D, Burla L. Psychische Gesundheit in Der Schweiz Monitoring 2012. https://www.npg-rsp.ch/fileadmin/npg-rsp/Themen-Bibliothek/obsan_psyGesundheit-3_2012.pdf
[2] Carvalho AF, Cavalcante JL, Castelo MS, Lima MCO. Augmentation strategies for treatment-resistant depression: A literature review. J Clin Pharm Ther 2007; 32(5): 415-428. doi:10.1111/J.1365-2710.2007.00846.X

[3] Gabriel FC, de Melo DO, Fráguas R, Leite-Santos NC, Mantovani da Silva RA, Ribeiro E. Pharmacological treatment of depression: A systematic review comparing clinical practice guideline recommendations. PLoS One 2020; 15(4):1-16. doi:10.1371/journal.pone.0231700

P-III-7

Characteristics of patients using anti-glaucoma therapy in the UK, 2000-2022

L. Velez-Nandayapa, L. Eggenschwiler, S.S. Jick, C.R. Meier, C. Becker

Basel Pharmacoepidemiology Unit, Division of Clinical Pharmacy and Epidemiology, Department of Pharmaceutical Sciences, University of Basel, 4031 Basel

Introduction: We previously reported on treatment rates for glaucoma or intraocular hypertension (IOH) in UK-based adults. There is little information on characteristics of such patients available.
Aims: We aimed to describe the population with anti-glaucoma therapy included in our previous study in terms of demographics, comorbidities and drug use, compared with a random sample of glaucoma-free patients from the base population.
Methods: Using data from the UK-based Clinical Practice Research Datalink (CPRD) Gold, we defined patients with anti-glaucoma therapy as individuals who received treatment for glaucoma or IOH (i.e., ≥ 2 prescriptions for anti-glaucoma medication, or glaucoma surgery) between 2000 and 2022. The earliest treatment prescription or surgery date served as the index date. Only patients 18 years or older and with ≥ 3 years of medical history in the database before the index date were included. Patients with anti-glaucoma therapy were matched 1:1 to glaucoma-free individuals from the CPRD population on age, sex, calendar time, general practice, and number of years of history in the CPRD prior to the index date. Characteristics of patients with and without anti-glaucoma treatment were compared using simple proportions. Differences between proportions were tested by Chi square tests.
Results: We found 120'804 patients with anti-glaucoma treatment, approximately half of whom were women. Mean age at start of anti-glaucoma treatment was 69.6 years. Women were slightly older than men at the start of treatment (mean age: 70.7 *vs.* 68.4 years). The two groups had largely comparable proportions of comorbidities. However, proportionately more patients with anti-glaucoma therapy had a diagnosis of diabetes (15.3% *vs.* 11.7%, p-value <0.01) or prescriptions for antidiabetic drugs (patients with 10 or more prescriptions: 9.3% *vs.* 7.4%, p-value <0.01). They also had more GP visits in the year prior to starting anti-glaucoma therapy. There were fewer current smokers among patients with anti-glaucoma therapy (12% *vs.* 13.9%, p-value <0.01). A diagnosis of dementia was less prevalent among individuals with anti-glaucoma therapy (1.2% *vs.* 2.1%, p-value <0.01). This observation may be explained by surveillance bias due to less vigorous screening for glaucoma in such patients.
Conclusions: Our analyses show that patients with and without anti-glaucoma treatment were similar though women started receiving anti-glaucoma therapy at a slightly later age than men and a higher proportion of patients with glaucoma had a diagnosis of diabetes compared to patients not treated for glaucoma or IOH. Glaucoma may be detected more often in patients with diabetes as such patients receive regular ophthalmologic check-ups.

Keywords: Glaucoma, patient population, medication, UK-based Clinical Practice Research Datalink (CPRD) Gold

P-III-8

Are potentially inappropriate mediations (PIMs) associated with rehospitalisation and death within three months? A systematic review and meta-analysis

I. Muzzarelli[1,2], V.I. Neumeier[2], A.R. Burch[2]

[1] *Institute of Pharmaceutical Sciences ETH, 8049 Zürich*
[2] *ZüriPharm AG, 8006 Zürich*

Introduction: The geriatric population is at an increased risk of receiving potentially inappropriate medications (PIMs). Currently, several screening tools are used in clinical practice to detect prescribed PIMs in geriatric patients and advise prescribers in optimizing drug therapies. This might reduce the risk of adverse events, unplanned rehospitalisation, or even death.
Aims: We aimed to assess the risk for rehospitalisation and mortality associated with detected PIMs by application of different screening tools.
Methods: Adhering to Cochrane standards, we conducted a systematic review with meta-analysis. We investigated the rehospitalisation and mortality rates within three months of patients aged 65 years and older with at least one prescribed PIM compared to patients without any PIMs. An explicit PIM detection tool must have been used to detect PIMs.
Results: Eight studies were included for the analysis of the outcome rehospitalisation and six for the outcome mortality. The applied PIM tools were the Beers criteria, Screening Tool for Older People's Prescriptions (STOPP), and EU(7)-PIM list. The analysis resulted in a pooled Odds Ratio (OR) of 1.47 (95% confidence interval (CI) 1.01–2.13, p=0.045) for rehospitalisation for patients with detected PIMs compared to patients without PIMs. This association was also statistically significant for the subgroup of studies, that only applied STOPP with a pooled effect estimate of 1.84 (95%-CI 1.08–3.12, p=0.024), but not for the application of Beers (OR 1.25; 95%-CI 0.86–1.81, p=0.235). Regarding the outcome mortality, no significant association between detected PIMs and mortality was found. Visual inspection of the forest plots suggested considerable heterogeneity between studies. The certainty of evidence was graded as very low for both outcomes.
Conclusions: Our findings indicate that even a single prescribed PIM leads to an increased odd of rehospitalisation within three months. We suggest the implementation of PIM tools in the medication evaluation of all patients aged 65 years and older in order to make prescribers aware of PIMs and enable drug therapy optimisation.

Keywords: Geriatrics, PIMs, Beers, STOPP, rehospitalisation

IV. CLINICAL PHARMACY / CLINICAL PHARMACOLOGY

P-IV-1

Identifying current practices and areas for improvement in medication management during care transition through an Interprofessional Collaboration Framework

L. Solh Dost[1,2], G. Maillard[1,2], E. Cardoso[1,2], M.P. Schneider[1,2]

[1]School of Pharmaceutical Sciences, University of Geneva, Geneva
[2]Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, Geneva

Introduction: Poor coordination and communication among healthcare professionals during care transitions can lead to medical errors, patient dissatisfaction, and hospitalization. Interprofessional collaboration is a promising strategy to enhance the quality and safety of care transition but needs to be better studied and implemented in practice.
Aims: This study aimed to describe the current state and areas for improvement of interprofessional collaboration regarding medication management during care transition from hospital to outpatient care.
Methods: This study, using qualitative methodology, was conducted through serial focus groups (FG) with patients, and in- and outpatient healthcare professionals, in Geneva, Switzerland. FG were audio-recorded, and transcribed verbatim. Data were analysed using thematic analysis and themes were then classified in the Canadian National Interprofessional Competency (CIHC) Framework.
Results: Twelve participants participated in four consecutive focus groups. Our study underlined current practices and areas for improvement in five of the six competency domains: 1. interprofessional communication, 2. patient partnership, 3. role clarification, 4. team functioning, 5. collaborative leadership and two macro-level additional improvements, 6. federating health professional associations and 7. improving policy support. Regarding current practices, healthcare professionals and patients highlighted the importance of patient-centred care and existing communication and collaboration practices between and within settings. However, healthcare professionals communicate reactively due to the lack of role clarity and collaborative leadership. Participants suggested measures for improving collaborations, such as shared information transmission channels, increased patient empowerment, and standardized discharge processes.
Conclusions: While existing practices emphasize communication and patient involvement, role clarity and collaborative leadership remain significant challenges. Healthcare professionals are motivated to work in collaboration, but policy support and coordinated efforts among healthcare entities are needed to ensure effective care, implement sustainable interventions, and improve patient outcomes during care transitions.

Keywords: Interprofessional collaboration, collaborative practice, continuity of patient care, hospital discharge, medication therapy management

References:

[1] Geese F, Schmitt KU. Interprofessional Collaboration in Complex Patient Care Transition: A Qualitative Multi-Perspective Analysis. Healthcare (Basel). 2023;11(3).
[2] Jones CD, Vu MB, O'Donnell CM, Anderson ME, Patel S, Wald HL, et al. A Failure to Communicate: A Qualitative Exploration of Care Coordination Between Hospitalists and Primary Care Providers Around Patient Hospitalizations. J Gen Intern Med. 2015;30(4):417-24.
[3] Solh Dost L, Gastaldi G, Dos Santos Mamed M, Schneider MP. Navigating outpatient care of patients with type 2 diabetes after hospital discharge - a qualitative longitudinal study. BMC Health Services Research. 2024;24(1):476.

P-IV-2

Medication reconciliation at hospital admission: findings and learnings from a pilot study on a Swiss general internal medicine ward

N. Schönenberger[1,2], L. Schnyder[1], A. Goetschi[1,2], T. Tritschler[3], C. Meyer-Massetti[1,4]

[1] Clinical Pharmacology and Toxicology, Department of General Internal Medicine, Inselspital - Bern University Hospital, 3010, Bern
[2] Graduate School for Health Sciences, University of Bern, 3012 Bern
[3] Department of General Internal Medicine, Inselspital – Bern University Hospital, 3010 Bern
[4] Institute of Primary Healthcare BIHAM, University of Bern, 3012 Bern

Introduction: Transitions between care settings increase medication risks [1]. Medication reconciliation at hospital admission mitigates errors and can therefore improve medication safety [2].
Aims: This study aimed to develop and pilot a medication reconciliation process for elderly, polymedicated patients upon hospital admission to a general internal medicine ward.
Methods: Eligible patients, aged ≥65 years, admitted via the emergency department to a designated general internal medicine ward with ≥7 medications, had their outpatient medication lists requested and reconciled by a pharmacist within 24 hours of admission. The admission medication lists were updated to reflect the most accurate medication history, and treating physicians were notified of discrepancies via progress notes. For each enrolled patient, we documented demographics, discrepancies, and involved medications. Furthermore, we assessed the resolution or acceptance of identified discrepancies and proposed interventions. The pilot study period was from November 13, 2023, to April 30, 2024.
Results: A total of 59 patients (mean age: 78.5 ± 6.7 years) with 412 discrepancies were included. Patients were taking an average of 11.0 ± 3.8 regular and 2.9 ± 3.2 as-needed medications. The mean number of discrepancies per patient was 7.0 ± 5.0. Common discrepancies included omitted medications (n = 195, 47%), dosing issues (n = 95, 23%), and incorrect or unknown manufacturer (n = 83, 20%). Of the proposed interventions (e.g., starting a medication), 70% were accepted by the treating physicians. Interventions involving regular medications had a higher acceptance rate (79%) compared to as-needed medications (60%). Frequently involved medications included agents acting on the renin-angiotensin system (n = 22), diuretics (n = 19), statins (n = 19), and paracetamol (n = 14). Medication reconciliation was not possible for 9 patients due to the absence of a primary care provider to provide an up-to-date medication list. Information transfer challenges, particularly incomplete or unclear medication lists, posed additional difficulties.
Conclusions: The implemented medication reconciliation process for elderly, polymedicated patients upon hospital admission predominantly identified omitted medications, with a notable acceptance rate of proposed interventions. Challenges in information transfer highlight the need for improved communication across interfaces of care to enhance medication reconciliation effectiveness and ultimately patient safety.

Keywords: Medication safety, medication reconciliation, transition of care, clinical pharmacy

References:

[1] Mekonnen AB et al. Pharmacy-led medication reconciliation programmes at hospital transitions: a systematic review and meta-analysis. J Clin Pharm Ther 2016; Apr;41(2):128-44
[2] Najafzadeh M et al. Economic value of pharmacist-led medication reconciliation for reducing medication errors after hospital discharge. Am J Manag Care 2016; Oct;22(10):654-661

P-IV-3

Quality indicators for the pharmacological management of chronic non-cancer pain in older adult patients: an integrative review

A.N. Goetschi[1,2], H. Verloo[3], N. Schönenberger[1,2], U. Wernli[1,2], C. Meyer-Massetti[1,4]

[1] Clinical Pharmacology and Toxicology, Department of General Internal Medicine, University Hospital Bern, 3010 Bern
[2] Graduate School for Health Sciences, University of Bern, 3012 Bern
[3] HES-SO Valais-Wallis, Department of Health, 1950 Sion, Valais
[4] Institute for Primary Health Care BIHAM, University of Bern, 3012 Bern

Introduction: Chronic non-cancer pain (CNCP) affects 28–88% of older adults. They also experience more medication-related problems and are more likely to receive insufficient pain therapy, impacting their quality of care. Quality indicators can be used to assess and improve the quality of their care.
Aims and objectives: This integrative review aimed to consolidate the scientific evidence on existing quality indicators for the pharmacological care of older adult patients with CNCP.
Methods: We systematically searched Medline and Embase via Ovid, CINAHL via EBSCO, the SCOPUS databases, and Google Scholar. We used backward citation searching to identify additional studies. Two reviewers independently screened the titles, abstracts and full texts. One extracted and charted the data and assessed the risk of bias; the second validated this.
Results: We screened 4,068 articles identified through the systematic search of databases and 2,774 articles identified via citation searching. Seventy-eight articles met our inclusion criteria and were retained for analysis; most were narrative reviews. We extracted 11 validated quality indicators and developed a further 243 based on quality criteria reported in the literature. Quality indicators were mentioned a median of once each, with 70 mentioned three times. Quality indicators covered different levels, from pharmacotherapy in general to particular substance groups and individual active substances. Many studies (i.e. narrative reviews) had an inherent high risk of bias.
Conclusions: This integrative review established a scientific basis for developing quality indicators for the pharmacological management of older adult patients with CNCP. The great variety of quality indicators reported suggests a complex and diverse patient population. After validation by experts and input from patients, these indicators could help improve the quality of care provided to older adult patients with CNCP and help focus specific interventions on vulnerable patients.

Keywords: Chronic pain, medication safety, older patients, quality in healthcare

P-IV-4

Digital healthcare services in community pharmacies: The Pneumoscope™ case study

C. Godot[1,2] C. Backes[1,2], C. Y. Gujer[1,2], N. Obegi[1,2], A. Perez[3], A. Gervaix[3], M. P. Schneider[1,2]

[1] School of Pharmaceutical Sciences, University of Geneva, Geneva
[2] Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, Geneva
[3] Division of Paediatric Emergency Medicine, Department of Women, Child and Adolescent, Geneva University Hospitals (HUG), University of Geneva, Geneva

Introduction: New healthcare devices based on artificial intelligence are spreading, opening up novel opportunities for innovative-health pharmaceutical services in decentralised primary care settings. The Pneumoscope™, a smart stethoscope, is an innovative digital tool that recognises lung sounds by associating them with specific respiratory pathologies using artificial intelligence. Effective triage and early detection methods implemented in primary health care settings such as community pharmacies (CPs) could help reduce pressure on hospital emergency departments, healthcare systems and save lives.
Aims: To understand how the Pneumoscope™ could be implemented in the healthcare system, this research aims at analysing the pharmacists' and patients' readiness to use this device for respiratory disease screening in CPs in the French speaking part of Switzerland.
Methods: A 2-stage exploratory cross-sectional study was conducted: 1) a qualitative analysis using semi-structured interviews and focus groups to understand the management of patients with respiratory problems and the pharmacists' readiness of using the Pneumoscope™ in their daily clinical practice; 2) a quantitative questionnaire on patients' readiness to use the Pneumoscope™ and on their level of confidence in AI in the healthcare domain. Data collection and analysis was carried out by two Master's students.
Results: Pharmacists see great potential in integrating e-health services into their daily clinical practice to improve their legitimacy in advanced triage and interprofessional collaboration in care coordination with physicians. Most patients were satisfied with the care they received in CPs, and patients readiness of the Pneumoscope™ was correlated with their level of confidence in AI ($p=0.0092$) and with their CP location ($p=0.0276$).
Conclusions: Digital devices such as the Pneumoscope™ enable pharmacists to use their skills and knowledge to enhance their clinical engagement in patient care and public health. Scientific evaluation of the Pneumoscope™'s effectiveness in interprofessional collaboration in primary care and role definition are essential steps towards recognition by health authorities and reimbursement by health insurers.

Keywords: Triage, Pneumoscope™, respiratory symptoms, COVID-19, artificial intelligence, community pharmacist, interprofessional collaboration, primary care

P-IV-5

PharmVisit: Interprofessional ward rounds with clinical pharmacist have the potential to improve medication safety and foster interprofessional collaboration

P. Stoyanova[1], N. Schönenberger[2,3], A. Goetschi[2,3], D. Müller, D. Bertschi, C. Meyer-Massetti[2,5]

[1] Department of Head, Musculoskeletal System and Ageing, Geriatric University Clinic, Inselspital – Bern University Hospital, 3010 Bern
[2] Clinical Pharmacology and Toxicology, Department of General Internal Medicine, Inselspital - Bern University Hospital, 3010 Bern
[3] Graduate School for Health Sciences, University of Bern, 3012 Bern
[4] Institute for Hospital Pharmacy, Inselspital – Bern University Hospital, 3010 Bern
[5] Institute of Primary Healthcare (BIHAM), University of Bern, 3012 Bern

Introduction: Elderly patients are often multimorbid, polymedicated, frail and cognitively impaired, all risk factors for medication-related problems. When admitted to inpatient care, interfaces to outpatient care pose an additional risk for information gaps leading to medication discrepancies and potential treatment errors. Interprofessional activities, like pharmacist-accompanied ward rounds, can improve medication safety. Therefore, the Geriatric University Clinic of the Bern University Hospital agreed to a pilot project, welcoming clinical pharmacists to ward rounds.
Aim: The aim of this study was to pilot test interprofessional ward rounds on a geriatric ward, assessing the impact on medication safety.
Methods: In this 6-months-pilot study, we implemented a weekly interprofessional ward round process (PharmVisit) with a focus on medication safety, starting in mid July 2023. A systematic medication analysis of all patients on a specific ward was executed one day prior to the PharmVisit by a clinical pharmacist, based on electronic patient information. The PharmVisits start with an exchange between physicians (mostly junior physicians), a clinical pharmacist and a regular nurse in the absence of the patient. Suggestions by clinical pharmacy are discussed before entering the patient rooms. Adjustments are either made directly in the IT system, declined or noted for further discussion with a senior physician. For each patient discussed during the PharmVisit, we documented the problem identified by the pharmacist, the reason for the intervention, drugs involved according to the ATC code, the intervention suggested, and the acceptance rate, using the GSASA classification system (www.gsasa.ch).
Results: Until mid January 2024, we saw 133 patients during 26 visits and discussed 294 potential interventions. The most common problem category was “risk due to medication therapy” (n=134, 46%), followed by “untreated indication” (n=47, 16%). Proposals related in particular to dose adjustments (19%, n=14 too low, n=42 too high) and additional medications (n=46, 16%). 151 suggestions (51%) were accepted, 41 (14%) rejected and 102 (35%) needed further clarification or were deferred to primary care providers in the discharge report.
Conclusions: With PharmVisit, we piloted an interprofessional ward round process to improve medication safety that will be implemented into daily practice in 2024. While acceptance rate of clinical pharmacy suggestions was comparable to other studies, increasing routine might improve collaboration. A survey is planned to improve acceptance and efficacy of the PharmVisit.

Keywords: Clinical pharmacy, interprofessional collaboration, ward rounds, medication safety

P-IV-6

Personalized adherence interventions with electronic monitoring: A case report on polypharmacy management in epilepsy

M.C. Sarbach[1], S. Ruegg[2], S.S. Allemann[1], I. Arnet[1]

[1] Pharmaceutical Care Research Group, Dept. Pharmaceutical Sciences, University of Basel, Klingelbergstrasse 50, 4056 Basel
[2] Neurology Clinic, University Hospital Basel, Petersgraben 4, 4031 Basel

Introduction: Epilepsy treatment is challenging due to the necessity of multiple daily regimens of anti-seizure medications (ASM). To determine adherence, therapeutic drug monitoring (TDM) is standard practice but falls short of the assessment of long-time adherence. Electronic monitoring enables a deeper understanding of adherence behavior and tailored interventions may be implemented. We present the case of a male patient, in his mid 30s, with symptomatic multifocal epilepsy and emotional instability since 1999, who manages his triple ASM treatment with pillboxes. He lives independently in a village and is working part-time in the city. Poor adherence led to an unstable course of disease with repeated severe seizure manifestations. In January 2021, his neurologist asked the Pharmaceutical Care Research Group (PCRG) in Basel, Switzerland, to support the patient with optimizing his ASM intake behavior.
Aims: Developing personalized adherence strategies for polypharmacy in epilepsy.
Methods: Baseline adherence was measured in 2021 with the recording card Time4Med™. The patient was then successively offered various adherence aids with electronic monitoring over several weeks: weekly punch cards (Pharmis®) with Time4Med™, and prefilled blister pouches (Medifilm®) in the electronic dispenser Medido®. Taking and timing adherence, correctly dosed days as well as drug holidays were calculated. An interview with open-ended questions on the usability and satisfaction of the devices was conducted in September 2023.
Results: Adherence with personal pillboxes was unsatisfactory leading to an unstable course of disease with recurrent seizures. The punch cards were too bulky so that the patient often left them at home, leading to 64% taking adherence, 63% timing adherence, 47% correctly dosed days and one drug holiday of five days (6 weeks in January to March 2022). Significant improvements in medication adherence metrics were observed with the transition to the electronic dispenser with 93% taking adherence, 90% timing adherence, 86% correctly dosed days (4 weeks in April to May 2023). No medication holiday was recorded during this period. No epileptic seizures were observed during both observational periods with electronic monitoring. Patient-reported feedback on device usability and satisfaction were in favor of the intervention with the electronic dispenser in promoting consistent medication intake and optimizing treatment outcomes.
Conclusions: The case highlights the complex nature of managing polytherapy in daily-life and the importance of personalized adherence interventions. Moreover, patient preferences were crucial in implementing the successful adherence strategy. Overall, the case underscores the value of interprofessional health care in optimizing treatment outcomes in epilepsy management.

Keywords: Adherence, intervention, electronic monitoring, epilepsy, polypharmacy, pharmaceutical care, interprofessional health care

V. MOLECULAR PHARMACOLOGY / MOLECULAR MEDICINE

P-V-1

From parasite to therapeutic: Expression, functional characterization, and therapeutic potential of the leech-derived complement inhibitor gigastasin

A. Blagojevic[1], P. Rüthemann[1,2,3], K. Widmer[1], D. Caglar[1], M. Tiric[1], M. Sovichan[1], R.B. Pouw[1], S. Rabbani[1], M.A. Lill[2], D. Ricklin[1]

[1] Molecular Pharmacy, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel
[2] Computational Pharmacy, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel
[3] SIB Swiss Institute of Bioinformatics, University of Basel, Basel, 4056 Basel

Introduction: The complement system is a crucial defense mechanism against pathogens, but its inappropriate activation can lead to clinical conditions such as ischemia-reperfusion injury (IRI) and autoimmune-hemolytic anemia (AIHA). As a result, therapeutic inhibition of the complement system has emerged as a promising approach. Parasitic host defense inhibitors, which often target early-response pathways, provide templates for new therapeutics. Gigastasin, a serine protease inhibitor from the giant Amazon leech, has been shown to inhibit both the classical (CP) and lectin (LP) pathways of complement activation [1, 2]. However, the complexity of this disulfide-rich protein has hindered its recombinant production in yields suitable for preclinical studies.
Aim: In this study, we describe the successful expression of gigastasin in a prokaryotic system and its potential for therapeutic applications.
Methods: Gigastasin was expressed in *E. coli* under optimized conditions and purified using affinity chromatography. The protein was characterized for identity, purity (SDS-PAGE, Western Blot, Mass Spectrometry), and thermal stability (nanoDSF). To enhance purity and remove endotoxins, the protein underwent reversed-phase HPLC processing. Endotoxin levels were measured under GMP conditions. The 6xHis-tag was removed using TEV protease to produce tag-free gigastasin. We tested target binding using SPR and assessed the protein's activity by monitoring direct C1s inhibition and complement activation in human, monkey, and mouse serum using CP and LP ELISAs.
Results: Our results demonstrate that recombinant gigastasin can be produced in *E. coli* with high yield and purity. The protein exhibited nanomolar affinity for C1s, but not proC1s, indicating its selectivity for the active enzyme. Gigastasin showed strong activity in substrate cleavage and human complement activation assays. Its activity in mouse and monkey serum suggests its translational potential. Additionally, gigastasin displayed significant thermal stability, allowing for purification via reversed-phase HPLC, lyophilization, and storage without losing activity. This step also significantly reduced endotoxin levels to those suitable for clinical applications.
Conclusion: Our findings position gigastasin as a promising preclinical candidate for developing therapeutics aimed at treating complement-mediated diseases, such as IRI during transplantation or stroke. Furthermore, gigastasin can serve as a molecular template for derivatives with enhanced affinity and/or selectivity profiles.

Keywords: Complement system, therapeutics, preclinical drug development

References:
[1] Sheppard PO et al. Patent Application No.: US 2002 / 0102256A1. 2002.
[2] Pang SS et al. The structural basis for complement inhibition by gigastasin, a protease inhibitor from the Giant Amazon Leech. J Immunol 2017; 199(11): 3883-3891.

P-V-2

squeezeMD: An open-source tool for protein interaction visualization and analysis

P. Rüthemann[1,2,3], **A. Blagojevic**[1], **M. Smieško**[2], **M. Lill**[2], **D. Ricklin**[1]

[1] *Molecular Pharmacy Group, University of Basel, 4056 Basel*
[2] *Computational Pharmacy Group, University of Basel, 4056 Basel*
[3] *SIB Swiss Institute of Bioinformatics, University of Basel, Basel, 4056 Basel*

Introduction: Analyzing protein-protein interactions is crucial for understanding binding events and fostering protein optimization tasks. Effective visualization tools are essential for these analyses. However, current tools are often proprietary, outdated, or difficult to use for end users. Bridging the gap between computational chemistry and biologists is therefore crucial. By providing user-friendly, standardized routines, we make complex analyses more accessible for everyone.
Aims: This study aims to present and evaluate squeezeMD [1], an open-source workflow designed to simplify and enhance the visualization and analysis of protein interaction surfaces through various computational means. The tool will be specifically benchmarked on serine proteases and their ligands.
Methods: The squeezeMD workflow integrates multiple modules, both published and unpublished, in particular for *in-silico* mutagenesis and molecular dynamics simulations. It excels in downstream trajectory analysis, providing detailed visualizations and quantifications of protein-protein interactions at the residue level using our advanced tool, Protein Scoring Pose Analysis (PoSco). PoSco distinguishing various biophysical interactions such as van der Waals forces, hydrophobic interactions, and water effects. The workflow automatically identifies and highlights key amino acids involved in the binding surface. Designed for flexibility, this tool operates seamlessly on both local computers and high-performance computing clusters, with smart allocation of GPU and CPU resources. It also generates a comprehensive report and saves results in a database, making result interpretation straightforward and accessible for the end user.
Results: squeezeMD has been utilized to investigate serine protease inhibitors, demonstrating its effectiveness in visualizing and analyzing interaction surfaces. These insights were particularly valuable for explaining empirical mutagenesis results.
Conclusions: squeezeMD is a powerful, user-friendly tool that enhances the analysis of protein interactions by combining multiple computational methods into a single platform. Its applicability extends beyond serine protease inhibitors to potentially all types of protein-protein interactions. This tool extends the molecular biologists' toolkit by streamlining the drug development process, offering guidance before the time-consuming protein engineering stages.

Keywords: Protein-protein interactions, protein visualization, molecular dynamics

Reference:
[1] https://github.com/pruethemann/squeezemd

P-V-3

Development of a platform for the expression of recombinant antibodies targeting complement proteins

F. Meyer, A. Blagojevic, E. Umnyakova, R.B. Pouw, S. Rabbani, D. Ricklin

Molecular Pharmacy, Department of Pharmaceutical Sciences, University of Basel, 4056 Basel

Introduction: Antibodies are an important tool in research as they can be used as markers or blockers in various experimental methods. However, the quality and consistency of polyclonal antibodies can vary, posing challenges for their reliable use. The single-chain variable fragment (scFv) is a format, where the two variable fragments of a monoclonal antibody are connected via a flexible linker. Coupling this scFv to the crystallizable fragment (Fc) of an IgG antibody produces a «minibody» that retains the key characteristics of an IgG antibody while facilitating recombinant expression and ensuring consistent quality.
Aim: The aim was to build a platform for the expression of minibodies, allowing for easy interchange of scFv and Fc regions to create diverse minibodies against complement proteins.
Methods: Genes encoding for various scFv against complement proteins were cloned into pFUSE-hIgG4-Fc2, producing constructs for different scFv-Fc minibodies. Transient transfection in Freestyle 293-F cells was done for protein expression. The resulting minibodies were purified using a Protein A column on an FPLC system, expression and purification were verified via SDS-PAGE and western blot.
Results: The results showed successful expression and purification of the minibodies and the presence of the Fc region was confirmed via western blot. Preliminary data indicate, that the minibodies can recognize their antigen in western blot and show inhibition in ELISA assays.
Conclusion: The scFv-Fc minibody format has been described before and facilitates the recombinant expression of antibodies compared to the full-length IgG antibodies. Based on the first results obtained, the inhibitory potential is retained for this antibody format. These antibodies are not used for therapeutic purposes but for laboratory applications, such as for assay development or controls. The platform will enable to produce different antibodies based on published protein sequences for various research purposes.

Keywords: Recombinant antibodies, protein expression

P-V-4

Breaking the boundaries of the clinical C3 inhibitor compstatin: Development of species-tolerant and long-acting analogs

S. Vogt[1], A. Lander[1], R. Aschwanden[2], C. Lamers[1,3], O. Schwardt[1], B.V. Geisbrecht[4], H. Meyer zu Schwabedissen[6], M. Lill[2], J.D. Lambris[5], M. Smiesko[2], D. Ricklin[1]

[1] Molecular Pharmacy Group, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
[2] Computational Pharmacy Group, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
[3] Institute of Drug Development, Medical Faculty, University of Leipzig, Germany
[4] Department of Biochemistry and Molecular Biophysics, Kansas State University, USA
[5] Department of Pathology and Laboratory Medicine, University of Pennsylvania, USA
[6] Biopharmacy Group, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel

Introduction: The compstatin family, known for its peptide-based complement C3 inhibitors, has undergone continuous optimization, expanding its role in biomedical research and therapeutic applications. Pegcetacoplan, a PEGylated compstatin derivative, has achieved FDA approval under the names Empaveli and Syfovre by Apellis. Despite these advancements, compstatin's specificity for human and primate C3 limits its use in many preclinical disease models, hindering translational studies. Additionally, there is potential for enhancing its pharmacokinetic properties.
Aim: The aim of this study is to develop new analogs of the clinically approved cyclic peptide, compstatin, that are active in rodent models and possess improved pharmacokinetic properties, thereby overcoming the limitations of species specificity and enhancing their potential for translational research and therapeutic applications.
Methods: By combining recent structural insights with experimental and homology models of mouse and rat C3b, we have identified the molecular determinants that dictate compstatin's species specificity. Our research indicates that this specificity is due to a limited number of drug-target contacts at the protein interface rather than steric hindrance. Utilizing *in-silico* redesign techniques, we introduced new peptide-protein interactions to achieve compatibility with both human and rodent targets.
Results: Evaluation of lead candidates based on target-binding kinetics and *in vitro* complement inhibition resulted in the identification of a promising derivative with effective affinity and inhibitory activity (IC_{50} = 13 µM) for rodent C3. The favorable pharmacokinetic properties of compstatin are driven by its strong binding to the C3 protein, which is highly abundant in blood (up to 1.3 g/L). To further improve compstatin's half-life, structure-guided optimization led to the development of new analogs with picomolar affinity for human C3b. Remarkably, adding a single methyl group increased the target residence time by 40-fold. Additionally, we are exploring novel strategies for extending the half-life by creating compstatin-conjugates that bind to circulating serum albumin using a C20 diacid.
Conclusion: In this study, we successfully developed new analogs of the cyclic peptide compstatin, which demonstrate activity in rodent models and possess enhanced pharmacokinetic properties. By leveraging structural insights and in-silico redesign, we identified molecular determinants of species specificity and introduced new peptide-protein interactions compatible with both human and rodent targets. The lead candidates exhibited effective affinity and inhibitory activity for rodent C3. These advancements address the limitations of compstatin's narrow species specificity and pharmacokinetics, paving the way for broader preclinical evaluations and potential therapeutic applications.
Keywords: Complement system, peptides, compstatin, pharmacokinetic, lipidation

References:
[1] Ricklin D et al. Nat Rev Nephrol 2017; 14: 26-47.
[2] Lamers C et al. Nat Commun 2022; 13: 5519.

P-V-5

A DNAzyme's catalytic comeback: Why two deleterious mutations are better than one

Z.V. Kürsteiner

Institute of Pharmaceutical Sciences, Department of Chemistry and Applied Biosciences, ETH Zurich, 8092 Zürich

Introduction: DNAzymes are catalytically active DNA sequences that are capable of cleaving complementary RNA substrates in presence of metal ion cofactors, without requiring any auxiliary proteins. This autonomy in action holds significant potential for therapeutic applications and distinguishes DNAzymes from protein-dependent oligonucleotides such as siRNAs. However, DNAzymes' dependence on toxic ions like Pb^{2+} limited their application to biosensing purposes. New hopes were brought by a second generation of DNAzymes that could operate effectively in the presence of physiologically relevant ions like Mg^{2+}. DNAzyme 8-17 [1] is one of the most studied DNAzymes for its therapeutic potential. It had been selected for its cleaving efficiency in presence of Mg^{2+} but its activity is turned out to be highest with Pb^{2+} [1, 2]. In fact, it was found that 8-17 is related over conserved residues with the DNAzyme GR5 [3], which cleaves exclusively with Pb^{2+} and is known for being one of the most sensitive and fastest-cleaving DNAzyme sequences [4, 5]. It could be shown by mutational studies that only two base positions could make the difference in cofactor selectivity between 8-17 and GR5 [5]. This points at the versatility of catalytic DNA sequences and highlights the hidden potential of first generation DNAzymes like GR5. To date, only two RNA-cleaving DNAzyme structures have been solved, one of which is 8-17 [6, 7]. To better understand metal ion selectivity, we need more data on DNAzyme structures especially for highly selective cofactor binders like GR5.

Aims: This work is an attempt to get some insight into GR5 secondary structure based on the known relationship between GR5 and 8-17. The 8-17 crystal structure shows an unexpected Watson-Crick-Franklin (WCF) base pair between the conserved residues G7 and C13 which are believed to correspond to G7 and C14 in GR5. A mutation of either G7 or C14 in GR5 would lead to almost complete loss of activity. Assuming a WCF base pair is present in GR5 as well, it is hypothesized that both residues can be mutated simultaneously to retain some of the activity.

Methods: The original GR5 and the GR5 mutants GR5-C8T (literature known, active), GR5-C14G (literature known, inactive) and GR5-G7CC14G (rescued C14 mutant?) were synthesized with a fluorescent tag. Cleavage was induced and monitored over time in order to determine the reaction rates.

Results: GR5 and the literature known mutants GR5-C8T and GR5-C14G behaved as expected. While the C8T mutation affects the activity only slightly, the C14G mutation results in almost complete loss of activity. The GR5-G7CC14G mutant shows improved activity compared with GR5-C14G, however, the activity is still clearly reduced compared to the original GR5.

Conclusions: Simultaneous mutation of both G7 and C14 in the GR5 DNAzyme to preserve a hypothetical WCF base pair results in improved activity compared to mutating C14 alone. This supports the hypothesis that G7 and C14 interact via WCF in GR5, similar to the interaction between G7 and C13 in 8-17. However, the significantly reduced activity of the GR5-G7CC14G mutant suggests that G7 and/or C14 are also involved in another crucial interaction. Referring back to the 8-17 structure, it is possible that G7 in GR5 plays a role in Pb2+ coordination.

Keywords: DNAzyme, GR5, mutational study, secondary structure, cofactor selectivity

References:

[1] Santoro SW & Joyce GF. Proc Natl Acad Sci USA 1997; 94: 4262–4266.
[2] Brown AK, Li J, Pavot CMB, Lu Y. Biochemistry 2003; 42: 7152–7161.
[3] Breaker RR & Joyce GF. Chem Biol 1994; 1: 223–229.
[4] Saran, R. & Liu, J. Inorg. Chem. Front. 3, 494–501 (2016).
[5] Ren W et al. ChemBioChem 2020; 21: 1293–1297.

[6] Liu H et al. Nat Commun 2017; 81 8: 1–10.
[7] Borggräfe J et al. Nature 2021; 6017891 601: 144–149.

VI. PHARMACOLOGY / BIOPHARMACY

P-VI-1

Tolerability of [^{161}Tb]Tb-SibuDAB, a novel radioligand for the treatment of prostate cancer, in healthy mice

K. Krieger[1,2,3], A.K. Mapanao[1], U. Köster[4], N.P. van der Meulen[1], R. Schibli[1,2], C. Müller[1,2]

[1] *Paul Scherrer Institute, 5232 Villigen PSI*
[2] *ETH Zürich D-CHAB, 8093 Zurich*
[3] *Inselspital Universitätsspital Bern, 3010 Bern*
[4] *Institut Laue-Langevin, 38000 Grenoble, France*

Introduction: PSMA-targeted radioligand therapy has been approved for the treatment of metastatic castration resistant prostate cancer. The Center for Radiopharmaceutical Sciences at Paul Scherrer Institute (PSI) developed an improved radioligand by modifying the ligand to increase plasma half-life and introducing the Auger-electron emitter terbium-161 for its superior dosimetric properties.
Aims: In light of the imminent dose escalation study of the phase-I trial PROGNOSTICS (NCT06343038), the toxicity of PSMA-targeted radioligand therapy using the novel radioligand [^{161}Tb]Tb-SibuDAB [1] was evaluated in healthy mice compared with clinically used radioligands.
Methods: FVB mice were injected with 15, 30 or 60 MBq [^{161}Tb]Tb-SibuDAB (1 nmol ligand, six animals per group). Reference groups were injected with 30 MBq [^{161}Tb]Tb-PSMA-I&T or [^{177}Lu]Lu-PSMA-I&T, while a vehicle control group received saline. Mice were weighed thrice weekly and assessed for predefined endpoint criteria. Complete blood counts were obtained from sublingual samples at days 10, 28, and 56 post injection (p.i.). Heparinized plasma was obtained from retrobulbar samples on day 56 p.i. and alkaline phosphatase activity, blood urea nitrogen, total bilirubin and serum albumin concentrations were measured. Animals were sacrificed and necropsied on day 56 p.i.
Results: No significant differences in body masses were noted between interventional, reference, and control groups. Significant reductions in erythrocyte and thrombocyte counts were found in mice that received 60 MBq [^{161}Tb]Tb-SibuDAB on day 10 p.i. Erythrocyte counts recovered by day 28 p.i., whereas modest reductions in thrombocyte counts persisted until day 56. Trends towards reduced thrombocyte counts were also found in 15 and 30 MBq [^{161}Tb]Tb-SibuDAB groups on day 10 p.i., but resolved by day 28 p.i. No significant differences in cell counts were found in reference groups receiving either [^{161}Tb]Tb- or [^{177}Lu]Lu-PSMA-I&T at any time. Significantly increased kidney masses were found upon necropsy in the 60 MBq [^{161}Tb]Tb-SibuDAB group. Statistically significant but numerically irrelevant declines in blood urea nitrogen in the 60 MBq [^{161}Tb]Tb-SibuDAB group and total bilirubin in the 15 MBq [^{161}Tb]Tb-SibuDAB group were noted. No significant differences were found for alkaline phosphatase activity and serum albumin relative to controls.
Conclusions: Findings of modest concern were limited to the mice injected with 60 MBq [^{161}Tb]Tb-SibuDAB. This activity would correspond to a 14.8 GBq human dose after body surface area conversion and is, thus, likely of little relevance towards further clinical translation. Histopathological results will be available shortly.

Keywords: Radioligand therapy, prostate-specific membrane antigen, auger emitter

Reference:
[1] Borgna F et al. Mol Pharm. 2022 Jul 4;19(7):2105-2114.

P-VI-2

Preclinical radionuclide therapy of neuroendocrine neoplasms using radiolabeled somatostatin analogues

A.K. Mapanao[1], S.D. Busslinger[1], A. Mehta[1], C. Favaretto[1], P.V. Grundler[1,2], Z. Talip[1,2], U. Köster[3], K. Johnston[4], R. Schibli[1,5], N.P. van der Meulen[1,2], and C. Müller[1,5]

[1] Center for Radiopharmaceutical Sciences ETH-PSI, Paul Scherrer Institute, Villigen-PSI
[2] Laboratory of Radiochemistry, Paul Scherrer Institute, Villigen-PSI
[3] Institut Laue-Langevin, Grenoble, France
[4] ISOLDE/CERN, Geneva
[5] Department of Chemistry and Applied Biosciences, ETH Zurich, Zurich

Introduction: Radionuclide therapy targeting the somatostatin receptor is currently employed in the clinics for the treatment of neuroendocrine neoplasms. Complete response is, nevertheless, rarely achieved, thereby demanding the development of improved radiopharmaceuticals. Herein, we propose the combination of somatostatin analogues with the alpha-particle-emitting terbium-149, a novel radionuclide which previously demonstrated promising therapeutic potential [1, 2].
Aim: The goal of this study was to compare two novel radiopharmaceuticals targeting the somatostatin receptor: the cell-internalizing [^{149}Tb]Tb-DOTATATE and the non-internalizing [^{149}Tb]Tb-DOTA-LM3 *in vitro* and in terms of safety and therapeutic efficacy in mice.
Methods: Cell viability after exposure to [^{149}Tb]Tb-DOTATATE and [^{149}Tb]Tb-DOTA-LM3 was evaluated in somatostatin receptor-positive AR42J tumor cells, a tumor cell line with exocrine and endocrine characteristics. DNA double-strand break formation in treated cells was visualized through immunofluorescence imaging. The therapeutic efficacy of [^{149}Tb]Tb-DOTATATE or [^{149}Tb]Tb-DOTA-LM3 at different activity levels (1 x 5 MBq and 2 x 5 MBq) was investigated in AR42J tumor-bearing mice. The tolerability of 2-fold higher cumulative activity than used for therapy was determined in immunocompetent mice without tumors. Blood cell counts were measured on Days 10, 28 and 56 and renal function evaluated at 10 weeks after injection of the radio-pharmaceutical.
Results: Tumor cell viability decreased in a radioactivity-dependent manner, with [^{149}Tb]Tb-DOTA-LM3 (EC_{50}: 0.5 kBq/mL) demonstrating a slightly higher potency than [^{149}Tb]Tb-DOTATATE (EC_{50}: 1.2 kBq/mL). The two radiopharmaceuticals introduced comparable and dense formation of DNA double-strand breaks, regardless of their respective subcellular localization. Treatment with a single injection of [^{149}Tb]Tb-DOTATATE or [^{149}Tb]Tb-DOTA-LM3 delayed tumor growth, leading to longer median survival times (16.5 and 19 days, respectively) compared to that of untreated mice (8 days). The injection of the radiopharmaceuticals on two consecutive days further extended the median survival of mice to 30 days for [^{149}Tb]Tb-DOTATATE and 29 days for [^{149}Tb]Tb-DOTA-LM3. The application of 20 MBq/mouse of either radiopharmaceutical was well tolerated, as indicated by the similar range of blood cell counts and comparable renal function between treated and untreated mice.
Conclusions: [^{149}Tb]Tb-DOTATATE and [^{149}Tb]Tb-DOTA-LM3 showed promising potential for radionuclide therapy of tumors that express the somatostatin receptor. The differences between the anti-tumor effects between [^{149}Tb]Tb-DOTATATE and [^{149}Tb]Tb-DOTA-LM3 were marginal, inferring that subcellular localization did not affect the treatment outcome. Due to the absence of critical hematological and renal adverse effects within the duration of the study, the quantity of activity administered for therapy could be increased to further enhance the therapeutic efficacy.
Keywords: Targeted radionuclide therapy, terbium-149, somatostatin analogues, DOTATATE, DOTA-LM3

References:
[1] Müller C et al. Pharmaceuticals 2014; 7: 353.
[2] Umbricht C et al. Sci Rep 2019; 8: 17800.

P-VI-3

Development of a novel transthyretin-binding PSMA radioligand characterized by an improved biodistribution profile

C. Vaccarin[1], A. K. Mapanao[1], L. M. Deberle[1], A. E. Becker[1], F. Borgna[1], G. Marzaro[2], R. Schibli [1,3], C. Müller[1]

[1] Center for Radiopharmaceutical Sciences ETH-PSI, Paul Scherrer Institute, 5232 Villigen-PSI
[2] Department of Pharmaceutical and Pharmacological Sciences, University of Padua, I-35131 Padua, Italy
[3] Department of Chemistry and Applied Biosciences, ETH Zurich, 8093 Zurich

Introduction: Radioligand therapy is an emerging modality for cancer treatment. This therapeutic concept harnesses the ability of radiolabeled compounds to deliver cytotoxic radiation doses directly to malignant cells while minimizing damage to the surrounding healthy tissues. Recently, [177Lu]Lu-PSMA-617 (Pluvicto™) has been approved for prostate-specific membrane antigen (PSMA)-positive metastatic castration-resistant prostate cancer [1].
Aims: Radioligand therapy is an emerging modality for cancer treatment. This therapeutic concept harnesses the ability of radiolabeled compounds to deliver cytotoxic radiation doses directly to malignant cells while minimizing damage to the surrounding healthy tissues. Recently, [^{177}Lu]Lu-PSMA-617 (Pluvicto™) has been approved for prostate-specific membrane antigen (PSMA)-positive metastatic castration-resistant prostate cancer [1]. The functionalization of [^{177}Lu]Lu-PSMA-617 with an albumin-binding entity has been extensively investigated in recent years to increase its tumor accumulation and, hence, therapeutic efficacy [2]. As suggested in one of our previous studies, binding to other plasma proteins present in the bloodstream at lower levels could also serve this purpose [3]. This study explored the potential use of a transthyretin-binding functionality to ameliorate the biological profile and tumor accumulation of PSMA radioligands.
Methods: The designed PSMA ligand equipped with a transthyretin binder (PSMA-TB-01) was synthesized via a convergent synthetic pathway involving a combination of in-solution and resin-supported methodologies. After radiolabeling with lutetium-177, the resulting [^{177}Lu]Lu-PSMA-TB-01 was evaluated *in vitro* using PSMA-positive PC-3 PIP cells and PSMA-negative PC-3 flu cells. Binding of the radiotracer to plasma proteins was measured using solutions of isolated transthyretin or serum albumin and human plasma samples. Biodistribution and single photon emission computed tomography (SPECT) studies were conducted in PC-3 PIP/ PC-3 flu xenografted mice.
Results: PSMA-TB-01 was obtained in a low yield (2%) but high chemical purity (>98%) after 16 synthetic steps. [^{177}Lu]Lu-PSMA-TB-01 showed high and specific uptake in PC-3 PIP cells (69 ± 3% after 4 h incubation) but not in PC-3 flu cells (<1%). [^{177}Lu]Lu-PSMA-TB-01 was characterized by a considerable binding to transthyretin and, to a lesser extent, also to human serum albumin. Biodistribution studies in tumor-bearing mice confirmed the enhanced blood retention of [^{177}Lu]Lu-PSMA-TB-01, which translated to a higher tumor uptake (69 ± 13% IA/g at 4 h p.i.) compared to that of [^{177}Lu]Lu-PSMA-617 (56 ± 8% IA/g at 4 h p.i.). The obtained biodistribution data were also confirmed by SPECT imaging.
Conclusions: Taken together, the results of this study confirm the feasibility of using a transthyretin binder to improve the biodistribution profile of a PSMA radioligand. This concept may also be expanded to other tumor-targeting radiopharmaceuticals in the future.

Keywords: Radioligand therapy, PSMA, transthyretin

References:

[1] Keam SJ. Lutetium Lu 177 Vipivotide Tetraxetan: First Approval. Mol Diagn Ther 2022; 26 : 467-475 ; doi:10.1007/s40291-022-00594-2.
[2] Lau J, Jacobson O, Niu G, Lin KS, Benard F, Chen X. Bench to Bedside: Albumin Binders for Improved Cancer Radioligand Therapies. Bioconjug Chem 2019; 30: 487-502; doi:10.1021/acs.bioconjchem.8b00919.

[3] Muller C, Farkas R, Borgna F, Schmid RM, Benesova M, Schibli R. Synthesis, Radiolabeling, and Characterization of Plasma Protein-Binding Ligands: Potential Tools for Modulation of the Pharmacokinetic

P-VI-4

Distribution and metabolic study in *SLCO2B1*$^{+/+}$ and *Slco2b1*$^{-/-}$ rats receiving chronic oral treatment of erlotinib

M. Rysz[1], N. Paloumpis[1], A. Schäfer[1], A. Kanthasan[1], N. Bianchi[1], I. Seibert[1], D. Ricklin[2], H. Meyer zu Schwabedissen[1]

[1] Biopharmacy Lab, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel
[2] Molecular Pharmacy, Department of Pharmaceutical Sciences, University of Basel, Basel

Introduction: The Organic Anion Transporting Polypeptide (OATP)2B1 is known to play a role in the hepatic uptake of its substrates of endogenous and exogenous origin [1]. One xenobiotic known to be transported by OATP2B1 is erlotinib. This EGF-receptor tyrosine kinase inhibitor is further known to be metabolized by cytochrome P450 enzymes where the CYP3A enzymes (CYP3A4 and CYP3A5) appear of major relevance. In the context of CYP-mediated metabolism, it was shown that erlotinib functions as a mechanism-based inhibitor resulting in irreversible inactivation of CYP3A enzymes [2].
Methods: Assuming that OATP2B1 may impact the accessibility of CYP enzymes for erlotinib in the liver, we conducted a study where we orally treated SLCO2B1$^{+/+}$ and Slco2b1 $^{-/-}$ Wistar rats with erlotinib for 8 consecutive days. During the study, the serum levels of erlotinib and its pharmacologically active metabolite OSI-420 were measured 1 hour after oral administration and immediately before the next dose was administered (trough levels). To measure the distribution of erlotinib in different tissues, organs were harvested (liver, small intestine and kidney) on the final day of the study. Finally, the livers were harvested for the preparation of microsomes applicable for functional CYP studies. The activity of CYP3A enzymes was determined with testosterone or midazolam, respectively.
Results: Using LC-MS/MS to quantify erlotinib and its major metabolite OSI-420 one hour after the last application, we observed a slight increase in OSI-420 levels in the serum of *SLCO2B1*$^{+/+}$ animals. Surprisingly, we also observed a rise in erlotinib and OSI-420 levels at the C_{trough} levels. From the tissues examined a significant increase was observed in the liver of *SLCO2B1* $^{+/+}$ animals for both erlotinib and OSI-420. Assessing the *ex vivo* activity of CYP3A-enzymes in liver microsomes isolated from the chronically treated *SLCO2B1*$^{+/+}$ and *Slco2b1* $^{-/-}$ rats, we detected no difference in the formation of 6-β-OH and 2-β-OH-testosterone from testosterone as determined by HPLC. Similarly, when using midazolam as a probe substrate of CYP3A, we observed no difference in the formation of 4'-OH midazolam, however a significant increase in the production of 1'-OH-midazolam in liver microsomes isolated from *SLCO2B1* $^{+/+}$ *rats* comparing to *Slco2b1* $^{-/-}$ rats (both were treated with erlotinib).
Conclusions: In conclusion, our *in vivo* results show that humanized (*SLCO2B1*$^{+/+}$) rats have increased erlotinib liver uptake after chronic treatment (and increased OSI-420 formation). Those changes were not observed in the kidneys and small intestine. The increased levels of both erlotinib and OSI-420 in *SLCO2B1*$^{+/+}$ rats at the C_{trough} levels suggest differences in compound handling that need to be further studied. Our CYP3A activity assays reveal a slight increase in the 1'OH-midazolam formation in *SLCO2B*$^{+/+}$ *rats*. Overall, after assessing the CYP3A activity levels using testosterone or midazolam in solvent-treated and erlotinib-treated rats' livers, we did not observe an inhibitory effect of erlotinib.

Keywords: Erlotinib, in-vivo rat model, drug transporters, SLCO2B1, cytochrome P450 activity

References:

[1] Kinzi J, Grube M, Meyer zu Schwabedissen HE. OATP2B1 - The underrated member of the organic anion transporting polypeptide family of drug transporters? Biochem Pharmacol 2021; 188: June 2021, 114534

[2] Li X, Kamenecka TM, Cameron MD. Cytochrome P450-mediated bioactivation of the epidermal growth factor receptor inhibitor erlotinib to a reactive electrophile. Drug Metab Dispos 2010; 38:1238-45

P-VI-5

Mission (im)possible: Target identification for natural products inhibiting oncogenic ERK and AKT signaling pathways in melanoma

M. Karpouchtsi[1], L. Dürr[1], M. Dobrzyński[2], S. Radetzki[3], T. Hell[1], M. Hamburger[1], J.P. von Kries[3], O. Pertz[2], R. Teufel[1], and E. Garo[1]

[1] *Department of Pharmaceutical Sciences, University of Basel, 4056 Basel*
[2] *Institute of Cell Biology, University of Bern, 3001 Bern*
[3] *Leibniz-Forschungsinstitut für Molekulare Pharmakologie, 13125 Berlin, Germany*

Introduction: Malignant melanoma is the deadliest type of skin cancer with unmatched mutation rates arising in both MAPK/ERK and PI3K/AKT signaling pathways. Although specific inhibitors of these critical pathways show spectacular initial results, most patients relapse within just a few months. Combination therapy can improve overall survival rates, but the currently available options are limited [1]. Novel specific inhibitors targeting oncogenic ERK and AKT signaling in melanoma are therefore urgently needed.
Aim: Target identification for natural products inhibiting oncogenic ERK/AKT signaling pathways
Method: Our in-house library of crude plant extracts was combined with an innovative high-content screening (HCS) assay that quantifies downstream inhibitory activity at ERK and AKT level. HPLC-based activity profiling of the active hits and subsequent targeted isolation of the bioactive constituents was performed [2]. To further explore the coverage of chemical space for such inhibitors, an additional screening campaign (EU-OPENSCREEN) on large pure compound libraries was launched by accessing high-throughput screening (HTS) pipelines.
Results & Conclusion: To this end, we screened 2,576 plant extracts and an additional 25,696 pure natural and synthetic compounds. A total of 47 active compounds were confirmed as downstream inhibitors of ERK and/or AKT with IC_{50} values in the low micromolar range. The current challenge aims towards target identification of the most promising hits. Several key kinases of the ERK-AKT network were produced through different cloning techniques and heterologous expression systems. Our strategy further includes the assessment of physical binding as well as enzymatic inhibition. First hints on specific targets for some of our newly discovered inhibitors will be presented.

Keywords: Target identification, natural products, MAPK/ERK and PI3K/AKT signaling, melanoma, high-content screening

References:
[1] Tanda ET, Vanni I, Boutros A, Andreotti V, Bruno W, Ghiorzo P, Spagnolo F. Current State of Target Treatment in BRAF Mutated Melanoma. Front. Mol Biosci 2020; 7: 154.
[2] Dürr L, Hell T, Dobrzyński M, Mattei A, John A, Augsburger N, Bradanini G, Reinhardt JK, Rossberg F, Drobnjakovic M, Gupta MP, Hamburger M, Pertz O, Garo E. High-Content Screening Pipeline for Natural Products Targeting Oncogenic Signaling in Melanoma. J Nat Prod 2022; 85: 1006-1017.

P-VI-6

Biochemical characterization of skin mitochondrial extracellular vesicles

F. Brigger[1], V. Mantella[1], D. Borghesi[2], S. Werner[2], J.-C. Leroux[1]

[1] Department of Chemistry and Applied Biosciences, ETH Zurich, 8093 Zurich
[2] Department of Biology, ETH Zurich, 8093 Zurich

Introduction: Extracellular vesicles (EVs) are small, natural vesicles released by various cells. They play a pivotal role in intercellular communication [1]. It was reported that the transfer of mitochondrial components, including proteins and mitochondrial DNA (mtDNA), via EVs can promote cellular homeostasis and prevent the death of damaged cells [2-4]. Recently, we found that EVs from wounds in healing-impaired diabetic mice contained elevated levels of mitochondrial proteins, suggesting potential implications in the wound healing process [5]. However, the transfer of mitochondrial elements through EVs (mit-EVs) and its implications for tissue homeostasis, repair, and disease is not well understood. Therefore, a comprehensive characterization of mit-EVs is important for determining their role in the context of wound healing.
Aims: This study aims at investigating the role of mitochondrial components in EVs derived from HaCaT keratinocytes, with a focus on understanding their implications in skin wound healing. Specifically, we aim to characterize EVs of different sizes isolated from HaCaT keratinocytes and to analyze their mitochondrial content.
Methods: EV subpopulations of different sizes were isolated from 3D cultures of skin cells (HaCaT keratinocytes) by ultracentrifugation and characterized according to MISEV guidelines [6]. Size and morphology were determined by nanoparticle tracking analysis, dynamic light scattering, transmission electron microscopy (TEM) and cryo-electron microscopy (cryo-EM), respectively. Protein profiling was conducted using western blotting. Further, subpopulations of EVs with different sizes were labeled with mitochondria-selective fluorescent probes and analyzed by nanoparticle flow cytometry (nanoFCM).
Results: Isolated EVs were enriched with specific EV marker proteins (i.e. CD63). Depending on the EV isolation procedure, average diameters of 112 nm and 148 nm could be distinguished. TEM displayed the typical cup-shaped morphology, while cryo-EM revealed the clear presence of a lipid bilayer. NanoFCM revealed a higher fraction of vesicles positive for mitochondrial components in larger EVs compared to smaller ones.
Conclusions: We isolated and characterized extracellular vesicles (EVs) of different sizes derived from HaCaT keratinocytes using an ultracentrifugation-based protocol. NanoFCM analysis revealed the presence of mitochondrial components within these EVs. Overall, this project represents a first effort in exploring the emerging field of mit-EVs, particularly in the context of skin wound healing.

This work was funded by ETH Zurich (SKINTEGRITY.CH project).

Keywords: Extracellular vesicles, intercellular transfer, mitochondria, skin, wound healing

References:
[1] Van Niel G et al. Nat Rev Mol Cell Biol 2018; 19: 213-228.
[2] Spees JL et al. Proc Natl Acad Sci USA 2006; 103: 1283-1288.
[3] Crewe C et al. Cell Metab 2021; 33: 1853-1868.e11.
[4] Phinney DG et al. Nat Commun.2015; 6: 8472.
[5] Krivitsky V et al. Adv Mater 2023; 35: 1-36.
[6] Théry C et al. J Extracell Vesicles 2018; 7: 1535750.

P-VI-7

Deciphering the influence of *SLCO2B1* on CYP3A-function: a rat study using midazolam

N. Paloumpis, M.A. Rysz, N. Bianchi, V. Taggi, I. Seibert, H.E. Meyer zu Schwabedissen

Biopharmacy, Department of Pharmaceutical Sciences, University of Basel, 4003 Basel

Introduction: The *SLCO2B1* gene encodes organic anion transporting polypeptide 2B1 (OATP2B1), essential for the cellular uptake and pharmacokinetics of various substrates. Despite the fact that OATP2B1 transports a variety of clinically used drugs, our understanding of its *in vivo* role is still limited. Recent findings from our laboratory showed increased expression of rCYP3A1, an orthologue of human CYP3A4, in rats expressing the human *SLCO2B1 (SLCO2B1*$^{+/+}$*)* compared to knockout (*rSlco2b1*$^{-/-}$) animals. Human CYP3A4 is known to be the most prevalent isoform of the CYP3A subfamily and is responsible for metabolizing approximately 50% of current drugs. Midazolam is not a substrate of OATP2B1 and the formation of its major metabolites, 1'-hydroxymidazolam (1'-OH MDZ) and 4-hydroxymidazolam (4-OH MDZ) is catalyzed by CYP3A4 in humans and by CYP3A1/A2 in rats.
Aim: In this study, we want to investigate the impact of OATP2B1 humanization on rCYP3A expression and activity by conducting a phenotyping study using midazolam as a probe drug in *SLCO2B1*$^{+/+}$ and *rSlco2b1*$^{-/-}$ rats.
Methods: This study evaluated the *in vivo* relevance of OATP2B1-mediated changes in rCYP3A activity by administering a single oral dose of midazolam and comparing its pharmacokinetics and metabolite formation (1'-OH MDZ and 4-OH MDZ) between the two genotypes. Additionally, liver microsomes from untreated *SLCO2B1*$^{+/+}$ and *rSlco2b1*$^{-/-}$ rats were exposed to midazolam to monitor metabolite formation using LC-MS/MS. Recombinant rCYP3A1/3A2 enzymes were used to investigate their contributions to midazolam hydroxylation.
Results: The concentration-time profiles showed that *rSlco2b1*$^{-/-}$ rats had lower serum concentrations of midazolam compared to *SLCO2B1*$^{+/+}$ rats during the initial phase (0-1 h), but comparable metabolite levels over time. The metabolite-to-parent drug ratio at 0.15h (t_{max}) indicated a significantly higher metabolic activity in *SLCO2B1*$^{+/+}$ rats. Pharmacokinetic analysis confirmed these findings regarding systemic exposure ($AUC_{0\rightarrow1h}$) and elimination rate (k_{el}). Interestingly, liver microsomes from knockout animals showed significantly higher formation of 1'-OH-MDZ and 4-OH-MDZ. Assays with recombinant enzymes indicated that both CYP3A1 and CYP3A2 metabolize midazolam, albeit to different extents.
Conclusions: Further investigation into the impact of humanization on midazolam metabolism by examining the differences between the two genotypes could provide deeper insights into OATP2B1's *in vivo* role.

Keywords: Midazolam, OATP2B1, rCYP3A1/A2, liver, metabolism

Reference:
Rysz M, Schäfer AM, Paloumpis N, Kinzi J, Brecht K, Seibert I, Schmidlin S, In-Albon K, Ricklin D, and Zu Schwabedissen HEM. Humanization of SLCO2B1 in rats increases rCYP3A1 protein expression but not the metabolism of erlotinib to OSI-420. J Pharmacol Exp Therap 2024; 389(1): 87-95.

P-VI-8

In vitro analysis of Cytochrome P450 induction and its impact on coproporphyrin levels

L. Potzel, J. Kinzi, A.M. Schäfer, I. Seibert, H.E. Meyer zu Schwabedissen

University of Basel, Department of Pharmaceutical Sciences, Biopharmacy, 4056 Basel

Introduction: Organic Anion Transporting Polypeptides (OATPs) are membrane proteins that facilitate the cellular uptake of endogenous molecules and xenobiotics. OATP1B1 and OATP1B3 are clinically relevant isoforms expressed in the liver which impact the pharmacokinetics of substrate drugs. Importantly, coproporphyrin (CP) I and CPIII, which are byproducts of heme synthesis, are considered suitable endogenous biomarkers for the evaluation of drug-drug interactions involving the hepatic uptake transporter OATP1B1. However, it is largely unclear whether CP levels can be impacted by other mechanisms besides the inhibition of OATP1B1. One mechanism could be the induction of the heme proteins cytochrome P450 (CYP). Indeed, increased CYP2B6 production may require an increase in heme synthesis, resulting in more corresponding byproducts, including CPI and CPIII.
Aims: This study focuses on the question whether metamizole and its active metabolite 4-methylaminoantipyrine (4-MAA) are interacting with OATP1B1 and whether they have an inducing effect on CYP2B6 and thereby affect heme biosynthesis and coproporphyrin levels *in vitro*.
Methods: To determine the impact of metamizole and 4-MAA on the OATP1B1-uptake of CPI and CPIII, we performed an *in vitro* transport experiment using MDCKII-OATP1B1 cells. Moreover, we performed a cell based reporter gene assay using HepG2 cells, to verify that 4-MAA and metamizole function directly as an activator of the Constitutive Androstane Receptor (CAR). In order to evaluate the impact of 4-MAA on expression of the heme protein CYP2B6, we conducted induction experiments in differentiated HepaRG cells.
Results: Transport inhibition studies suggest no significant impact of neither metamizole nor its metabolite 4-MAA on the OATP1B1-mediated cellular accumulation of coproporphyrins. In addition, we observed transactivation of CYP2B6 by CAR in HepG2 cell-based reporter gene assays when treated with metamizole and 4-MAA. Expression of CYP2B6 was monitored by real-time PCR and Western blot analysis in HepaRG cells treated with 4-MAA.
Conclusion: In summary, we report an effect of metamizole and its active metabolite 4-MAA on CYP2B6 expression, whereas no effect on the OATP1B1 transporter was observed. Future studies *in vivo* are warranted to determine whether the intake of metamizole leads to increased CP levels.

Keywords: OATP1B1, coproporphyrins, biomarker, metamizole, 4-MAA, heme proteins, CYP2B6, CYP3A4, CAR

P-VI-9

Investigating the dual functionality of branched tetrameric peptides for their applications in chronic wound treatment

N. Vavilthota[1], G. Babuccu[1], J.W. Drijfhout[2], R.A. Cordfunke[2], M. Riool[1], S.A.J. Zaat[1]

[1] Department of Medical Microbiology & Infection Prevention, Amsterdam institute for Infection and Immunity, Amsterdam UMC, University of Amsterdam, Meibergdreef 9, 1105 AZ Amsterdam, The Netherlands
[2] Department of Immunology, Leiden University Medical Center, 2300 RC Leiden, The Netherlands

Introduction: Chronic wounds pose a significant clinical challenge due to their persistent nature and susceptibility to infections. These wounds often harbour multi-drug resistant pathogens, complicating treatment and prolonging healing times. Traditional antimicrobial therapies are increasingly ineffective, necessitating the development of novel therapeutic agents with both antimicrobial and wound healing properties. Lactoferricin, a peptide derived from bovine lactoferrin, has shown promise in addressing these challenges due to its broad-spectrum antimicrobial activity and ability to promote wound healing. In the recent years, branched peptides with higher copy number of motifs have shown to have superior antimicrobial activity compared to the monomeric motif.

Aim: To investigate the antimicrobial and wound healing functions of four bovine lactoferricin-derived synthetic tetrameric peptides: LAP-47, LAP-48, LAP-49 and LAP-50. Here the peptide candidates were composed of four RRWQWR monomeric motifs linked together using the connector region composed of lysine (LAP-47) or diaminopropionic acid (LAP-48) or diamino-propionic acid oxalic acid (LAP-49) and D-lysine (LAP-50) and cysteine residues with a spacer to avoid steric hindrance. In the present study, these branched peptide candidates were characterized for their dual functio-nality along with testing for improvement in the activity with the modifications.

Methods: First, epithelial and endothelial cell line, human keratinocyte (HaCaT) and human umbilical vein endothelial cell were used to demonstrate and screen the wound healing potential of the four peptides in a 2D scratch and tube formation assay respectively. For immunomodulatory studies, the peptides were tested for bacterial endotoxins neutralisation in macrophages. Best performing peptide candidates were tested for anti-fungal and broad-spectrum anti-bacterial activity against the ESKAPE panel. Lastly, the *ex vivo* human skin wound infection model was used to confirm the antimicrobial activity of the peptides.

Results: All peptides showed potent antimicrobial activity against gram positive and gram negative bacteria (0.47-0.94 μM). In the cell migration after wounding and in angiogenesis assay, peptides LAP-47 and LAP-48 showed significant cell migration ($P < 0.001$) and dense tube formation compared to the controls and were selected for further characterizations. LAP-47 and LAP-48 also showed a wide therapeutic window compared to LAP-49 and LAP-50 from the cytotoxicity evaluations. Additionally, these peptides were able to diminish bacterial endotoxins induced tumor necrosis factor-α levels from macrophages indicating potential anti-inflammatory effects. Macro-phages pretreated with peptides were able to augment intracellular bacterial killing resulting in infection clearance and wound healing in difficult to heal wounds. In the *ex vivo* human skin wound model, the tetramer LAP-48 effectively reduced *S. aureus* bacterial load in a concentration-dependent manner ($P < 0.01$) and showed better activity compared to LAP-47.

Conclusions: These findings underscore the potential of branched tetrameric peptides as multi-functional therapeutic agents for treating hard to heal infected chronic wounds. Further, the modifications in the connector region would have likely altered the properties of the peptides, which was seen in improved antimicrobial and wound-healing properties in LAP-48. Future studies will focus on stability tests and additional immunomodulatory functions involved in wound healing for these peptides.

Keywords: Chronic wound treatment, novel antimicrobials, antimicrobial peptides and wound healing.

P-VI-10

Modulation of autophagy against multiple myeloma and use of *C. elegans* as a study tool

J. Vériène-Salerno[1,2,3], **M. Cuendet**[1,2,3]

[1] *School of Pharmaceutical Sciences, University of Geneva, 1211 Geneva*
[2] *Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, 1211 Geneva*
[3] *Translational Research Center in Oncohaematology, University of Geneva, 1211 Geneva*

Introduction: Autophagy is a process used by cells to recycle molecules essential for their survival under unfavorable conditions. Tumor cells are surrounded by a hostile, acidic, and nutrient-poor environment, and autophagy is essential for their survival. However, autophagy is also needed for immune cells to fight tumor cells [1]. Simply blocking autophagy to stop tumor growth is often not sufficient, and it is important to understand the autophagy characteristic features between healthy and tumor cell types [2].
Aim: The aim of this study is to understand autophagy processes in multiple myeloma (MM), a hematological cancer with uncontrolled plasma cells proliferation, in order to have a more targeted approach in treating this disease.
Methods: Here, we show that autophagy modulation in MM should be followed with specific markers. Using various tools such as western blot and confocal microscopy, we were able to monitor the regulation of autophagy over time in multiple myeloma cell lines, in samples from MM patients, and in organoids containing MM cells. In parallel, the nematode *C. elegans* and especially an mCherry::GFP::LGG-1 strain is used to study this mechanism in a more complex context and to follow autophagy *in vivo* over time, since this process is extremely well conserved across species.
Results: P62 is a protein degraded upon autophagy activation that is often used as a marker. However, in MM cells, p62 did not represent a good marker since it behaved differently from one MM cell line to another. In addition, we have identified an antiproliferative compound that displayed a higher activity in MM cells compared to solid cancer cell types and appeared to regulate autophagy. Our work aims at understanding this mechanism of action.
Conclusion: While many studies focus on autophagy modulation in various diseases, the success rate of autophagy-related clinical trials would certainly be higher with a better understanding of autophagy characteristic features depending on the cell types. The present study may also lead to new treatment to fight MM.

Keywords: Multiple myeloma, autophagy, organoids, *C. elegans*, P62.

References:

[1] Xia H, Green DR, Zou W. Autophagy in tumour immunity and therapy. Nat Rev Cancer 2021; 21: 281–297.
[2] Zhan L, Li J, Wei B. Autophagy therapeutics: preclinical basis and initial clinical studies. Cancer Chemother Pharmacol 20218; 82: 923–934.

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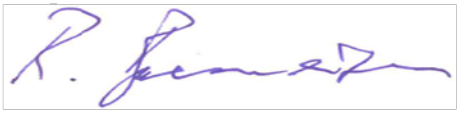

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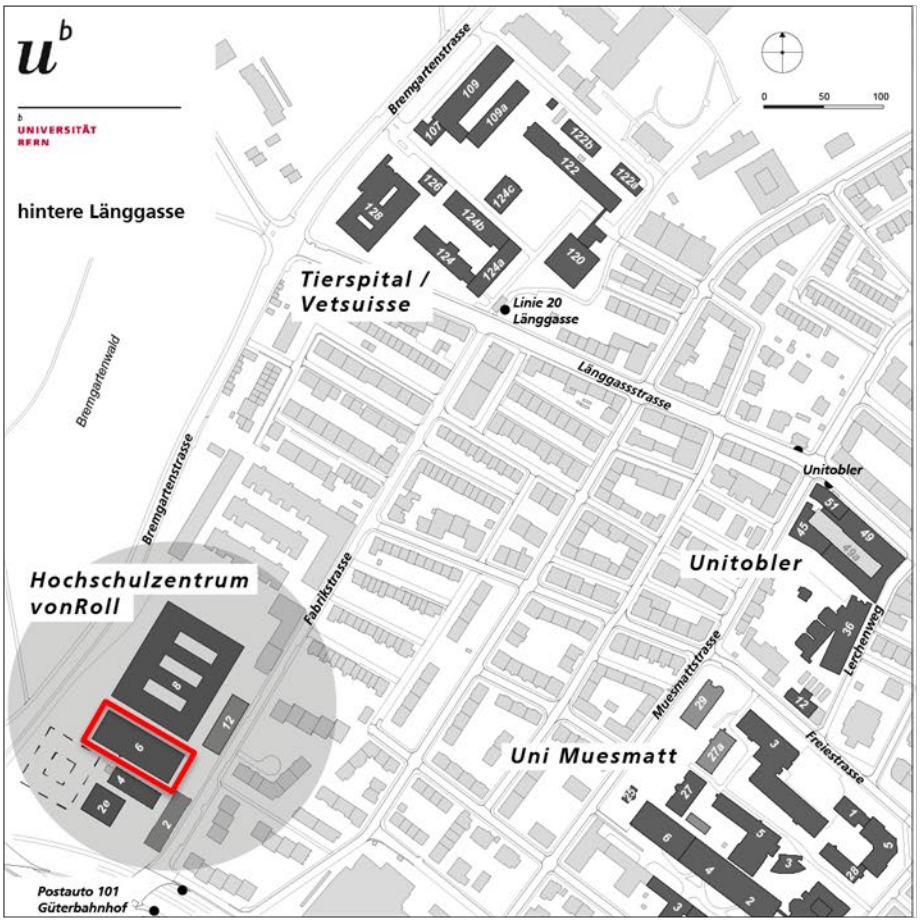

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