

# 2024

# HH-CORES SUMMER SCHOOL PRUGRAM 2-7 SEPTEMBER 2024

PRESENTED BY Helmholtz-Health Core Facilities Network

e-mail



**ADDRESS:** Venusberg Campus 1/99 53127











# WELCOME

elcome to the Helmholtz-Health Cores summer school. Our summer school is designed exclusively for PhD students in the biomedical sector and offers an opportunity to learn cutting-edge research technologies and methods, collaborate with peers, and engage with leading experts in the field.

With a focus on interdisciplinary learning, our program is divided into four distinct tracks, each tailored to provide comprehensive insights into key areas of biomedical research. You will find a common track for everybody dedicated to FAIR use of scientific data and you can choose one of the other tracks focusing in Advanced Light Microscopy, Laboratory Automation, and Image and data analysis. While you will work in groups focusing on a specific project, all tracks work synergistically and will interact with each other's. We will select 30 motivated Ph.D. students that will be assigned to the three tracks (10 for each track). We are looking forward to host you to our summer school and help you to expand your skill set, and propel your research forward. Join us and enjoy a full week of high-end learning at the DZNE in Bonn.



# MULTIMODAL IMAGING, ARE SET TO BRIDGE THE GAPS BETWEEN SUB-CELLULAR AND ORGANISM Imaging.

# **ABOUT THE SUMMER SCHOOL**

The Helmholtz-Health Core Facilities network represents the intersection of cutting-edge research and collaborative innovation. Each Helmholtz Health center offers specialized central technology platforms known as Core Facilities (CF).

Core Facilities provide a centralized infrastructure that ensures fast, convenient, and affordable access to state-of-the-art technologies and services. By democratizing expensive and complex technologies, CFs enable scientists to solve challenging problems more efficiently. These facilities are operated by highly skilled staff with the expertise needed to maximize their utility. Continuous method development within the CFs and ongoing exchange with the scientific community keep them at the forefront of science.

Our first summer school is aiming to offer the know-how present in our CF to young scientists. The Summer School will be hosted at DZNE Bonn and is designed exclusively for PhD students in the biomedical sector. It will offer an opportunity to learn cutting-edge research technologies and methods, collaborate with peers, and engage with leading experts in the field.

With a focus on interdisciplinary learning, our program is divided into four distinct tracks, each tailored to provide comprehensive insights into key areas of biomedical research. You will find a common track for everybody dedicated to FAIR use of scientific data and you can choose one of the other tracks focusing in:

Track 1: Advanced	Track 2: Advanced	Track 3: Image and
Light Microscopy	Laboratory Automation	Data Analysis
You will learn about three	You will learn advanced	You will cover three topics
major topics of advanced	laboratory automation for	related to image and data
light microscopy:	drug screening:	analysis.
<ol> <li>Large tissue imaging</li> <li>High-resolution deep- tissue imaging</li> <li>Imaging subcellular structures by super- resolution microscopy.</li> </ol>	<ol> <li>Automated liquid handling techniques</li> <li>Automated multiparametric HCS assay processing</li> <li>Identification of most promising hit candidates.</li> </ol>	<ol> <li>Analysis of high-content screening images and data</li> <li>Building a neural network for cell type classification</li> <li>Segmentation of spheroid SUSHI imaging.</li> </ol>

# HOW TO APPLY



# SUBMIT YOUR APPLICATION

Showcase your academic background, research interests, and motivation to attend by writing a compelling application letter. Submit it together with your CV. Choose your priority for track you will like to attend. **Deadline for application submission is 22nd July 2024.** 



# **SELECTION PROCESS**

Our summer school is **limited to 30 students**. We will select the best candidates and hope you will be part of the team! A waiting list will be created, and if any of the first 30 candidates do not register, you will be informed and given the opportunity to join the summer school.



The first 30 candidates will be informed by the **26th of July** and you will have time to register and pay the fee till the **2nd of August**.

Registration fee only 500 € to be paid after successfull selection You can indicate your preference for Track 1 through Track 3. However, please note that expressing a preference does not guarantee placement in your chosen track.

# **ABOUT THE LOCATION**

The HH-Cores Summer school will be hosted at the DZNE in Bonn. DZNE was founded in 2009 as a member of the Helmholtz Association and the first member of the German Centers for Health Research (DZG). Today, it consists of ten sites – Berlin, Bonn, Dresden, Göttingen, Magdeburg, Munich, Rostock/Greifswald, Tübingen, UIm and Witten – and pools expertise distributed nationwide within a single research institute. More than 1,200 professionals collaborate to understand the causes of diseases of the nervous system and to find novel approaches for effective prevention, therapy, and patient care. Worldwide, DZNE is one of the largest research institutes dedicated to these topic. It is funded by the German Federal Ministry of Education and Research (BMBF) and by the federal states in which DZNE sites are located. DZNE is involved in the implementation of the National Dementia Strategy for Germany.

Research at DZNE aims at preventing neurodegeneration and significantly improve the quality of life of affected individuals. Thus, DZNE dedicates its efforts towards all aspects of neurodegenerative diseases – from molecular mechanisms to health care and patient care. In order to cover this wide range of topics, DZNE pursues an interdisciplinary scientific strategy that includes various interlinked areas of research: DZNE scientists work together across disciplines and sites, so that new findings can be rapidly transferred into practical application. Within this "translational" approach, DZNE collaborates with universities, university hospitals, research institutes and research-driven companies – both in Germany and worldwide.

# **SOME INFORMATION ABOUT ACCOMODATION**

### We organized a call-off quota until August 4 with the:

Intercity Hotel Quantiusstraße 22 53115 Bonn Website

Requested rooms can be canceled free of charge until 6 pm on the day of arrival. Please contact the organizer to obtain the keyword for the contingent call-off.

Reservations can be made by telephone on +49 228 926181513 or by email to **reservations.bonn@intercityhotel.com** 



# **OUR SPEAKERS**

# PETER HORVATH

Director Institute of Biochemsitry, BRC, Szeged Principal Investigator at AI for Health HMGU

Peter research focus on the intersection of biology, engineering and computer science, and combines wet-lab and light microscopy with image analysis and machine learning methods.

# VALENTIN NÄGERL

Professor: University of Bordeaux

Valentin is professor of neuroscience and bio-imaging at the University of Bordeaux, where he directs the master's program in cellular bio-imaging and heads a research team focusing on the nano-structural mechanisms of neural plasticity by applying novel in vivo super-resolution microscopy approaches

# **OUR SPEAKERS**



# **ANIKA STEFFEN**

Head of the Central Facility for Microscopy HZI - Braunschweig

Anika is heading the central facility for micrsocopy at the HZI in Braunschweig. Her expertise cover live cell imaging, in particular TIRF microscopy, spinning disk confocal, SIM (structured illumination microscopy) and photomanipulation techniques, and operates different high end microscopes at the HZI.

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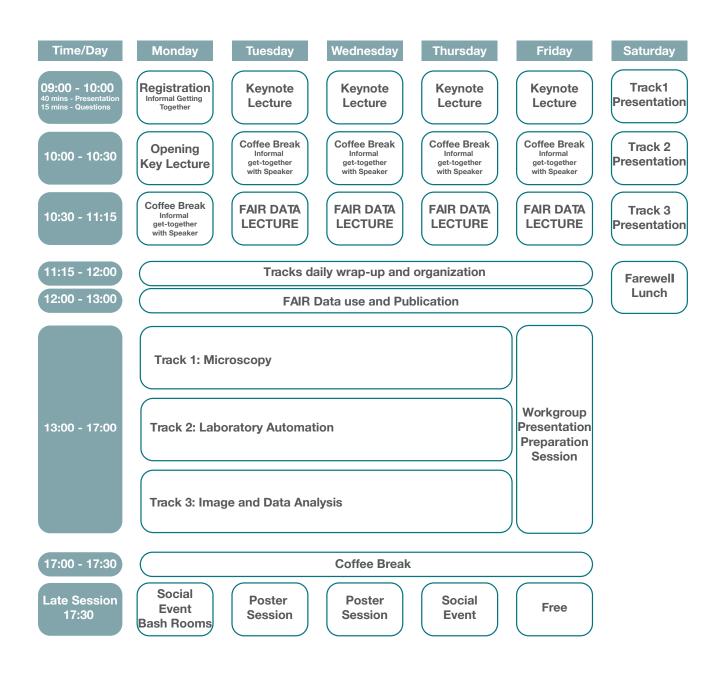


# **STEFAN PRECHTL**

Senior Scientific Consultant at Cellima

Stefan founded CELLIMA in 2021. He specializes in high-content imaging and has expertise in high-throughput screening, automated image analysis and machine learning. Rely on Stefan and CELLIMA to advance scientific discovery with unparalleled expertise, reliability and efficiency.

# DAILY SCHEDULE AT A GLANCE



# SHARED TRACK: FAIR DATA AND PUBLICATION

In the open science landscape, FAIR principles for scientific data management and stewardship are now the gold standard. This series of lectures endeavors to introduce you to the principles of FAIR, which are: Findable, Accessible, Interoperable and Reusable and provide you with information to inform your own research data management planning to ensure they conform to this standard.

## WHAT WILL I LEARN?

You will learn about the background underpinning FAIR data in the context of an open science

landscape and why it is important. You will understand the principles of FAIR data, understand basics of FAIR data with regard to sensitive information. You will learn about best practices for FAIR data in order to ensure that your data corresponds to FAIR principles. Finally, you will learn about data management planning in order to provide structure for implementing your own data management plan.

# **COURSE CONTENT**

Monday: Groups kick off Tuesday: FAIR Data: Why FAIR? What is FAIR? Wednesday: FAIR Data for Sensitive Data Thursday: Lecture: FAIR Data: Interoperability & Reusability? Make it so! Friday: Data Management Planning

# **RECOMMENDED PREPARATORY READING:**

- 1. Beyan, O, Choudhury, A., van Soest, J., Kohlbacher, O., Zimmermann, L., Stenzhorn,
- 2. H., Karim, M.R., Dumontier, M., Decker, S., Santos, L.O.B.S & Dekker, A. (2020). A distributed analytics on sensitive medical data: The personal health train. Data Intelligence, 2(1-2). https://doi.org/10.1162/dint\_a\_00032
- 3. Barker, M., Chue Hong, N.P., Katz, D.S, Lamprecht, A-L., Martinez-Ortiz, C.
- 4. Psomopoulos, F., Harrow, J., Castro, L.J., Gruenpeter, M., Martinez, P.A. &
- 5. Honeyman, T. (2022). Introducing the FAIR principles for research software. Scientific Data 9(622). https://doi.org/10.1038/s41597-022-01710-x
- 6. European Commission, Directorate-General for Research and Innovation. (2018).

- 7. Turning FAIR into reality Final report and action plan from the European Commission expert group on FAIR data, Publications Office. https://data.europa.eu/doi/10.2777/1524
- 8. Foster, E. and Deardorff, A. (2017). Open science framework. Journal of the Medical
- 9. Library Association, 105(2): 203-206. https://doi.org/10.5195/jmla.2017.88
- 10. GO FAIR. (n.d.) FAIR principles. Retrieved May 15, 2024, from
- 11. https://www.go-fair.org/fair-principles/
- 12. Jacobsen, A., Azevedo, R.M., Juty, N., Batista, D., Coles, S., Cornet, R., Courtot, M.,
- Crosas, M, Dumontier, M., Evelo, C.T., Goble, C., Guizzardi, G., Hansen, K.K., Hasnain, A., Hettne, K., Heringa, J., Hooft, R.W.W., Imming, M.,...Schultes, E. (2020). FAIR principles: Interpretations and implementation considerations. Data
- 14. Intelligence, 2(1-2). https://doi.org/10.1162/dint\_r\_00024
- 15. OpenAIRE. (2020.) How to make your data FAIR. Retrieved May 15, 2024 from
- 16. https://www.openaire.eu/how-to-make-your-data-fair
- 17. Wilkinson, M.D., Dumontier, M., Aalbersberg, I.J., Appleton, G., Axton, M., Baak, A.,
- Blomberg, N., Boiten, J-W., Santos, L.B.S., Bourne, P.E., Bouwman, J., Brookes, A.J., Clark, T., Crosas, M., Dillo, I., Dumon, O., Edmunds, S., Evelo, C.T., Finkers, R., ...Mons, B. (2016). The FAIR guiding principles for scientific data management and stewardship. Sci Data, 3(160018). https:// doi.org/10.1038/sdata.2016.18

# TRACK 1: ADVANCED LIGHT MICROSCOPY METHODS

Light sheet microscopy and other strategies for imaging entire organs

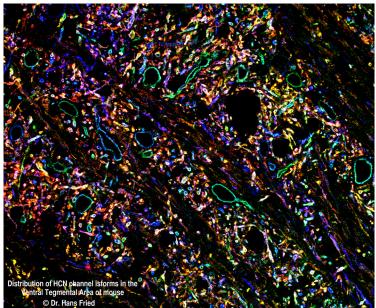
## **INTRODUCTION**

The Track1 course will provide the participants with three major topics of advanced light microscopy: 1) Large tissue imaging, 2) High-resolution deep-tissue imaging, and 3) Imaging subcellular structures by super-resolution microscopy.

# LARGE TISSUE IMAGING:

This section of the workshop focuses on light-sheet microscopy and methods of tissue clearing.

We will introduce you to the principles of light sheet microscopy, showcasing its advantages over traditional methods, including reduced phototoxicity and accelerated image acquisition. You will also learn about the theory of light-sheet microscopy and you will gain hands-on experience in how to optimize imaging conditions to achieve high-resolution, three-dimensional images of complete organs.



### WHAT WILL I LEARN?

You will explore the practical applications of light sheet microscopy through

comparisons with other microscopy techniques. You will focus the day on various tissue-clearing methods, understanding the potential for transparent imaging without the need for sectioning. In addition, we will investigate the potential of light sheet microscopy for correlating diverse imaging technologies including magnetic resonance imaging, multiphoton microscopy, and super-resolution microscopy. By the end of the workshop, participants will be equipped with the knowledge and skills to begin the implementation of light sheet microscopy and tissue clearing techniques in their research projects.

## **COURSE CONTENT**

Wrap up session (morning session): Principles of Light Sheet Microscopy and clearing **Workshop (5h afternoon sessions):** 

- Protocols for clearing sections and how to select to best one (40 min)

- Sample mounting for light sheet microscopy using custom printed holders (40 min)
- Imaging with the Light-Sheet 7 from Zeiss (140 min)
- Calibrations and alignment to improve light sheet imaging and hands on imaging with various large tissue specimen
- Using light sheet data to correlate various imaging techniques (40 min)
- Compare light sheet with alternative imaging methods (40 min)

### **RECOMMENDED PREPARATORY READING:**

- 1. The Art of Tissue Clearing. Jacques Paysan, 2021. Microscopy and Analysis. Wiley.
- Light Sheet Fluorescence Microscopy: A Review. Peter A. Santi. J Histochem Cytochem. 2011 Feb;59(2):129-38. doi: 10.1369/0022155410394857.
- 3. Light sheet microscopy of living or cleared specimens. P.J. Keller and H-U. Dodt. Current Opinion in Neurobiology. 2012 Feb;22(1):138-43. doi: 10.1016/j.conb.2011.08.003.

## **HIGH-RESOLUTION DEEP-TISSUE IMAGING:**

This section of the workshop focuses on fundamentals and applications of multiphoton microscopy and how multiphoton microscopy improves imaging within live animals. We will discuss and show the different optical components of a multiphoton microscope and how to optimize them to enhance penetration depth and resolution. We will investigate applications of multiphoton microscopy in neuroscience, with a spotlight on imaging delicate neuronal structures and capturing neuronal activity.

### WHAT WILL I LEARN?

You will learn the theory of two- and three- photon microscopy and familiarize yourself with various hardware components like pre-chirp compensator, adaptive optics components, telescopes, and non descanned detectors. You will gain hands-on experience in aligning lasers and strategically placing mirrors along the beam path for optimal laser alignment. You will build up knowledge about suitable dyes and fluorescent proteins. By the end of the workshop you will be able to navigate the complexities of multiphoton microscopy and understand challenges of intravital microscopy in neuroscience.

## **COURSE CONTENT:**

Wrap up session (morning session): Principles in Multiphoton Microscopy

#### Workshop (5h afternoon sessions):

- Theory and hands on tutorial how to set up beam paths (30 min)
- How to align a laser or how to walk a beam (30 min)
- Group delay compensation: practical guide to optimize fluorescence output (30 min)
- - Modules for controlling laser power and laser power calibration (30 min)
- - Optimizing telescope settings for deep tissue imaging (30 min)
- Adaptive optics modules and hands on modification of the wave front (50 min)
- Imaging various specimen: from fixed specimen to awake mouse imaging. (100 min). A practical introduction

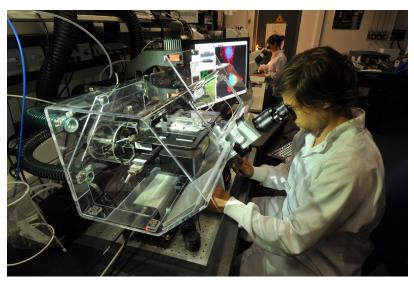
## **RECOMMENDED PREPARATORY READING:**

- Two-photon laser scanning fluorescence microscopy. Denk W, Strickler JH, Webb WW. Science. 1990 Apr 6;248(4951):73-6. doi: 10.1126/science.2321027. PMID: 2321027
- 2. Learning Zone Scientifica homepage: Multiphoton Imaging A Deep Look Series. https://www. scientifica.uk.com/learning-zone

## **IMAGING SUBCELLULAR STRUCTURES:**

This section of the workshop will introduce super-resolution microscopy techniques to increase the resolution below the diffraction limit of light. Specifically, we will introduce the theory of Airyscan microscopy and Stimulated Emission Depletion (STED) microscopy. Furthermore, handson sessions will allow attendees to operate state-of-the-art Airyscan and STED microscopes on-site to image subcellular components in various specimen, and how to further increase resolution with image processing.

### WHAT WILL I LEARN?



You will learn the principles of Airyscan and STED microscopy, including their unique mechanisms of enhancing resolution. During hands-on sessions, you will investigate possibilities to optimize the setup to increase resolution and imaging depth for biological applications. Various approaches of image processing to further increase the resolution will be explored. Furthermore, you will learn how to apply the increased resolution for co-localization analysis. By the end

of the workshop, you will be able to evaluate the potential of super-resolution microscopy and how to implement it in your scientific projects.

## **COURSE CONTENT:**

Wrap up session (morning session): Principles of super-resolution microscopy Workshop (5h afternoon sessions):

- Theory and hands on tutorial on Stimulated Emission Depletion Microscopy (120 min)
- Optimization of the depletion donut
- Imaging with various specimen
- Increasing the resolution via iterative deconvolution approaches
- Theory and hands on tutorial on Airyscan microscopy (120 min)
- Optimization of the Airy detector and improving co-registration accuracy
- Imaging with various specimen
- Analyzing Images with Airy processing and Joint Deconvolution algorithms

- Strategies to increase penetration depth in super-resolution microscopy (60 min)

### **RECOMMENDED PREPARATORY READING:**

- Super-resolution microscopy demystified. Schermelleh L, Ferrand A, Huser T, Eggeling C, Sauer M, Biehlmaier O, Drummen GPC. Nat Cell Biol. 2019 Jan;21(1):72-84. doi: 10.1038/s41556-018-0251-8. Epub 2019 Jan 2. PMID: 30602772
- 2. The Airyscan detector from ZEISS: confocal imaging with improved signal-to-noise ratio and super-resolution. Joseph H. Nature Methods volume 12, pages i–ii (2015).
- 3. Stimulated Emission Depletion Microscopy. Blom H, Widengren J. Chem Rev. 2017 Jun 14;117(11):7377-7427. doi: 10.1021/acs.chemrev.6b00653. Epub 2017 Mar 6. PMID: 28262022

# **IMAGING 3D-SPHEROIDS:**

This section of the workshop will introduce 3D cell culture and how to image them with the example of cellular spheroids. 3D cell cultures like organoids and spheroids are commonly used to bridge the gap between animal models and classical 2D cell culture systems. Yet, they have very unique requirements to cultivate, stain, and image them. We will introduce 3D cell culture and explore suitable imaging technologies to acquire entire cell spheroids.

### WHAT WILL I LEARN?

You will apply methods for 3D cell culture to cultivate spheroids in different sizes. The team will explore the challenges of 3D fluorescence stainings. You will learn during hands-on sessions the advantages of several, for 3D cell culture commonly used, imaging technologies including confocal microscopy, spinning disk microscopy, super-resolution microscopy, light-sheet microscopy, and two-photon microscopy.

## **COURSE CONTENT:**

Wrap up session (morning session): 3D cell culture systems Workshop (5h afternoon sessions):

- Various spheroids and their cell culture conditions. (60 min)
- Imaging Spheroids: comparing several imaging strategies
- spinning disk microscopy (60 min)
- confocal and super-resolution microscopy (60 min)
- multiphoton microscopy (60 min)
- light-sheet microscopy (60 min)

# TRACK 2: ADVANCED LABORATORY AUTOMATION

High-Content Screening (HCS) for anti-inflammatory Drugs – Chasing the novel Lead

## **INTRODUCTION**

The Track-2 course will provide the participants with three major topics of advanced laboratory automation for drug screening: 1) Automated liquid handling techniques, 2) Automated multiparametric HCS assay processing, and 3) Identification of most promising hit candidates.

### WHAT WILL I LEARN?

#### Day-1: Automated liquid handling techniques (4 h)

The participants of this course will learn how to utilize advanced techniques for liquid handling

in 384 well assay plate format miniaturize and to upscale throughput in а screening campaign. The instrumentation and corresponding software are "state-of-the-art" and are used worldwide in pharmaceutical and advanced academical screening labs. At the end of this course, the team will get all tools in hand to perform an advanced multiparametric HCS assay using a primary human heterotypical cell model.



### **COURSE CONTENT:**

- Standardization of heterotypical PBMC cell culture (30 min)
- Liquid handling in 384 well microtiter plate dispensing vs. pipetting (45 min)
- Non-contact liquid handling in nl quantity using acoustic energy (60 min)
- Accuracy control for liquid handling devices basic, but most important (45 min)
- Spotting of 300 unknown drugs in defined assay format (60 min)

Results of the day:

- "Ready-to-use" drug plates for day 2
- Protocols for automated liquid transfers for day 2

#### Day-2: Test of 300 drugs using a multiparametric inflammation assay 4 h

The teams will perform an entire primary HCS screen, in which they test unknown drugs for the capacity to inhibit innate immune response and inflammasome activation in PBMCs. They will learn how to combine homogenous and single cell assay approaches for detailed characterization

of drug effects. We will show them why and how to set up a standardized meta-data protocol for automated assay analysis.

Course content:

- 5-Step assay plate processing using automated liquid handling (180 min)
- Set up a standardized meta-data protocol for automated assay analysis (60 min) Results of the day:
- Processed assay plates
- Standardized tracking of meta-data

#### Day-3: Automated post-processing of screening plates 4 h

The participants of this course will learn how to do multiplexed immunofluorescence staining in 384well format and why different pipetting techniques will affect assay performance. They will get introduced into miniaturized HTRF assay technology to measure cytokine concentrations in the supernatant of tested wells. They will learn automated image acquisition using our CV8000 high-throughput microscope. Finally, the teams will get insight how to set up automated image analysis protocols to quantify drug-dependent cellular phenotypes.

Course content:

- Immunofluorescence staining for image-based single cell quantification (90 min)
- Measure cytokine concentration using homogenous HTRF assay- (60 min)
- Automated image acquisition using CV8000 (90 min)

Results of the day:

- Image data sets to analyze drug-dependent cellular phenotypes

#### Day-4: Identification of most promising hit series 4 h

In the final course the team will learn how to analyze huge standardized data sets of collected measurements and meta-data. They will team up with the data analysis experts from Track-3 and do together data analysis to identify the most promising hit series.

Course content:

- Measure cytokine concentration using homogenous HTRF assay-II (60 min)
- Standardized data handling of different measurements for assay analysis (60 min)
- Team up with a team of Track-3 (Image and Data Analysis) (60 min)
- Hit Identification using multiparametric features (60 min)

Results of the day

- Standardized assay results (data and meta-data)
- Identification of the most promising hit series

## **RECOMMENDED PREPARATORY READING:**

- 1. eBook, The Assay Guidance Manual, Eli Lilly & Company and the National Center for Advancing Translational Sciences
- 2. Buchser W, Collins M, Garyantes T, et al., Assay Development Guidelines for Image-Based High Content Screening, High Content Analysis and High Content Imaging.
- 3. https://www.ncbi.nlm.nih.gov/books/NBK100913/

4.

- 5. Josephine Blersch, Birgit Kurkowsky, Anja Meyer-Berhorn, Agnieszka K. Grabowska, Eva Feidt, Ellen Junglas, Wera Roth, Dominik Stappert, Armin Kübelbeck, Philip Denner, Eugenio Fava, An ex vivo human model for safety assessment of immunotoxicity of engineered nanomaterials
- 6. bioRxiv 2023.06.29.547008; doi: https://doi.org/10.1101/2023.06.29.547008

- 7.
- 8. McManus RM, Latz E. NLRP3 inflammasome signalling in Alzheimer's disease. Neuropharmacology. 2024 Jul 1;252:109941. doi: 10.1016/j.neuropharm.2024.109941. Epub 2024 Mar 31. PMID: 38565393.
- 9.
- Kelley N, Jeltema D, Duan Y, He Y. The NLRP3 Inflammasome: An Overview of Mechanisms of Activation and Regulation. Int J Mol Sci. 2019 Jul 6;20(13):3328. doi: 10.3390/ijms20133328. PMID: 31284572; PMCID: PMC6651423.

# TRACK 3: IMAGE AND DATA ANALYSIS Introduction

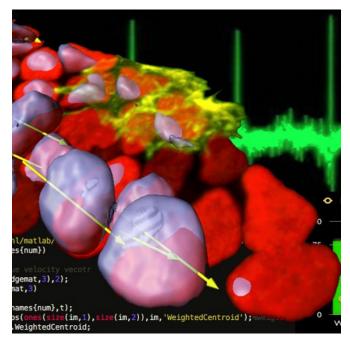
In the Track-3 course, we will cover three topics related to image and data analysis. 1) Analysis of high-content screening images and data, 2) building a neural network for cell type classification, 3) analysis of SUSHI imaging of spheroids.

### DAYS 1&2: HIGH-CONTENT SCREENING ANALYSIS

In this workshop, we will explore image and data analysis in the context of a HCS compound screening. We will analyze multichannel fluorescence high-content images, extract relevant data and set up a workflow to process the data generated by the automated microscope. We will then explore the generated data and build a multi-variate analysis to identify compounds performing best for given criteria.

### WHAT WILL I LEARN?

You will learn how to approach the analysis of large amounts of data generated by a screening facility. You will process and structure the raw data, generate primary and



secondary features and set up a multi-variate analysis to classify all used chemical compounds. We will then sort the compounds according to given criteria and identify the best ones in regards to the given question.

## **COURSE CONTENT:**

Talk (morning session): Introduction to HCS images, data and automated image analysis

- Analysis of multi-channel fluorescence images
- Preprocessing and structuring large amounts of data
- Multi-variate data analysis
- Presentation of results

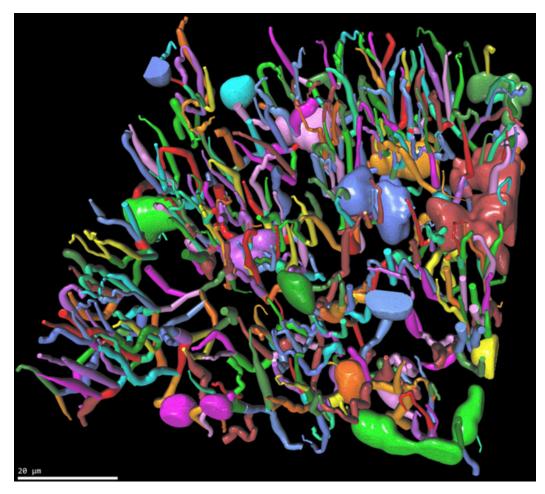
## DAYS 384: ANALYSIS OF SUSHI IMAGING OF SPHEROIDS

In this workshop, we will analyze a 3D stack of images of a spheroid recorded via state-of-the-art SUSHI imaging (SUper-resolution SHadow Imaging). You will segment individual cells and look at

subcellular structures of individual cells of the multi-cell spheroid. Then we will analyze changes over time in the segmented regions.

### WHAT WILL I LEARN?

You will learn how to analyze a 3D time-lapse stack of images of a 3-dimensional multicellular object. To do this, you will learn to segment cells and subcellular structures and track changes over time.



## **COURSE CONTENT:**

Talk (morning session): Introduction to SUSHI imaging, z-stacks and time-lapse imaging

- Analysis of SUSHI imaging
- Cell and subcellular object segmentation
- Analysis of time-lapse imaging

## DAY 4: NEURAL NETWORK CELL TYPE CLASSIFIER

In this workshop, we will build a neural network classifier to distinguish three cell types based on fluorescence images of their nuclei. Images of fluorescent nucleus markers of monocytes, T-Cells and B-Cells will be used as label data to train the network. The finished classifier will then be applied to a new set of images and the results will be analyzed.

## WHAT WILL I LEARN?