



7th Annual Mass Cytometry Summit

Friday, April 27, 2018

Hilton Prague Old Town

Grand Ballroom Dvořák

A background image showing a cluster of cells, likely from a mass cytometry experiment, with various colors and textures. The cells are overlaid with a white grid.

							76 Cd	77 In
					77 Ir		78 Pt	79 Au
					57 La		58 Ce	59 Pr
							64 Gd	65 Tb
65 Tb	66 Dy	67 Ho	68 Er	69 Tm	70 Yb	71 Lu		

CONTENTS

5	Welcome
6	Agenda
9	Presentation abstracts and speakers
20	CYTO Fluidigm Commercial Tutorials

VÍTEJTE!

Welcome to Prague and the 7th Annual Mass Cytometry Summit.

On behalf of Fluidigm, thank you for taking time to spend the day with us and colleagues from around the world to discuss the exciting new insights made by the mass cytometry community this past year. It is great to have so many familiar faces here and exciting to meet the many first-time Summit attendees as well.

The advancements made with mass cytometry in immunology, immunotherapy and cancer research over the last few years have been truly impressive, opening the door to the development of better diagnostics and more effective therapies in the future. With the nearly exponential rise in publications since 2013, mass cytometry is well-recognized around the world as an essential tool for high-parameter analysis of cell phenotyping and function.

Today we look forward to learning from scientific and technical leaders sharing their latest research findings, applications and data analysis solutions for mass cytometry and Imaging Mass Cytometry™. In turn, we will share recent improvements to the mass cytometry workflow and introduce an exciting new research product for comprehensive human immune monitoring. Over the course of the day, we will ensure that there is plenty of time for the dynamic discussions, open exchanges and collaborations that emerge during every annual summit.

We are incredibly optimistic about the future of health care. In collaboration with you, our singular goal is to unlock the full potential of high-parameter cell and tissue analysis.

Sincerely,

A handwritten signature in black ink that reads "S. Christopher Linthwaite". The signature is written in a cursive, flowing style.

Chris Linthwaite

President and CEO, Fluidigm

AGENDA

Welcome

8:00

Registration

Continental Breakfast

8:45

Welcoming Remarks

Chris Linthwaite, President and CEO, Fluidigm

Translating Mass Cytometry

9:00

Using Cell Signaling Pathways to Assess Immune Cell Dysfunction

Stephen Rapecki, PhD, UCB Celltech

9:30

Epigenomic-Guided Mass Cytometry Profiling Reveals Disease-Specific Features of Exhausted CD8 T Cells

Bertram Bengsch, MD, PhD, University Medical Center Freiburg

10:00

Coffee Break

10:15

Mapping the Maternal Immune Response to Normal and Pathological Pregnancies

Brice Gaudillière, MD, PhD, Stanford University School of Medicine

10:45

Advancing Human Systems Immunology

Petter Brodin, MD, PhD, Karolinska University Hospital

Novel Applications

11:15

Unraveling Human Microglial Heterogeneity by Multiplexed Mass Cytometry

Chotima Böttcher, PhD, Charité, University Medicine Berlin

11:35

Mass Cytometry for Immune Profiling in Chronic Inflammation

Henrik Mei, PhD, DRFZ Berlin, a Leibniz Institute

AGENDA

11:55 **Advancing Human Immune Monitoring with an Innovative Panel Kit and Data Analysis Approach**
Greg Stelzer, PhD, Fluidigm

12:15 **Lunch**

Best Practices

1:15 **New Best Practice Protocols for Sample Preparation and Acquisition**
Dmitry Bandura, PhD, Fluidigm

1:30 **Preparing for Clinical Trials in Bergen: From Sample Collection to Data Analysis**
Sonia Gavasso, PhD, Haukeland University Hospital, University of Bergen

1:50 **CD Maps: Tracking Antigen Expression During Human Thymic Development**
Tomáš Kalina, MD, PhD, Charles University in Prague

Data Analysis

2:10 **Powering Multi-Omic Translational Studies with Mass Cytometry**
Nima Aghaeepour, PhD, Stanford University School of Medicine

2:30 **Multidimensional View of the Innate Immune System in HIV-1-Infected Individuals**
Antonio Cosma, PhD, CEA, Université Paris Sud

2:50 **Optimizing Computer-Assisted Analysis of Immune Response to High-Fat Diet in Mouse Adipose Tissue and Liver**
Olivier Molendí-Coste, PhD, INSERM, Institut Pasteur de Lille

3:15 **Coffee Break**

AGENDA

- 3:30** **Imaging Mass Cytometry**
Empowering an Imaging Revolution
in Tissue Pathology
Mina Lakshman, PhD, Fluidigm
- 3:45** **Analysis of Tissue Ecosystems in Health**
and Disease by Imaging Mass Cytometry
Bernd Bodenmiller, PhD, University of Zurich
- 4:15** **Deep Phenotyping of Colorectal Cancer by**
High-Dimensional Mass Cytometry Reveals
Tumor-Specific Immune Landscapes
Frits Koning, PhD, Leiden University Medical Center
- 4:45** **Panel Discussion: Slice or Dice?**
When to Image and When to Suspend Samples
for CyTOF Analysis
Frits Koning, PhD, Leiden University Medical Center
(Moderator)
Bernd Bodenmiller, PhD, University of Zurich (Panelist)
Jared Burks, PhD, University of Texas MD Anderson
Cancer Center (Panelist)
Susanne Heck, PhD, Guy's and St Thomas' NHS
Foundation Trust and King's College London (Panelist)
Jonathan M. Irish, PhD, Vanderbilt University (Panelist)
- 5:30** **Closing Remarks**
Rob Ellis, PhD, Fluidigm
- 6:30** **Dinner Reception**
Francouzská Restaurace Art Nouveau

ABSTRACTS AND SPEAKERS

Using Cell Signaling Pathways to Assess Immune Cell Dysfunction



Stephen Rapecki, PhD

Director Functional Screening
UCB Celltech, Slough, UK

Profiling human immune responses by measurement of intracellular signaling pathways has already been established as a powerful technology using traditional fluorochrome-based flow cytometry.

This presentation will demonstrate the potential for mass cytometry to be a superior technology to traditional flow cytometry for the measurement of intracellular signaling. Using antibody panels labeled in-house, we have established a panel of phospho-specific antibodies covering up to 50 different signaling pathways. I will show that measuring both baseline and perturbed signaling responses in blood immune cells can reveal single-cell responses that help describe complex biology. Using informatics tools, I will show how these readouts can be used to monitor immune status, which may have benefit in both health and disease.

ABSTRACTS AND SPEAKERS

Epigenomic-Guided Mass Cytometry Profiling Reveals Disease-Specific Features of Exhausted CD8 T Cells



Bertram Bengsch, MD, PhD

Group Leader, Department of Internal Medicine II
University Medical Center Freiburg

Exhausted CD8 T cells (TEX) are a distinct T cell lineage and immunotherapy target in chronic infection and cancer but poorly defined in human diseases. We therefore developed a transcriptomic-

and epigenetic-guided mass cytometry approach based on exhaustion-specific genes that identified at least nine distinct TEX clusters in HIV and lung cancer by phenotypic, functional and transcription factor and inhibitory receptor co-expression patterns. An exhaustion severity metric was integrated with high-dimensional phenotypes to define TEX clusters that were shared across chronic infection and cancer or enriched in either disease; linked to disease severity; and changed with HIV therapy. These data also identified combinatorial patterns of immunotherapy targets on different TEX clusters. Profiling TEX heterogeneity has implications for immune monitoring and immunomodulation in chronic infections, autoimmunity and cancer.

ABSTRACTS AND SPEAKERS

Mapping the Maternal Immune Response to Normal and Pathological Pregnancies



Brice Gaudillière, MD, PhD

Assistant Professor, Department of Anesthesiology
Stanford University School of Medicine

The maintenance of pregnancy relies on a finely tuned immune balance between tolerance to the fetal allograft and protective mechanisms against invading pathogens. Combining a mass cytometry approach with a novel cell-signaling-based elastic net algorithm, we have identified communities of immunological events in maternal blood that precisely time gestation in a term pregnancy. Here, I will present an overview of ongoing research aiming at determining whether the timing of maternal immunological events during pregnancy, or the "immune clock" of pregnancy, is disrupted in pathological pregnancies such as preeclampsia and preterm labor.

Advancing Human Systems Immunology



Petter Brodin, MD, PhD

Assistant Professor, Science for Life Laboratory
Karolinska Institutet and Department of Neonatology
Karolinska University Hospital, Stockholm

I will present my group's recently developed technological advances in analyzing small-volume blood samples from newborn children by mass cytometry as well as the biological understanding learned from these analyses. I will also present recent developments the group has made in understanding human immune system variation and systems-level regulation using large-scale mass cytometry profiling in large numbers of samples. Finally, my talk will describe methods for integrating mass cytometry data with other data types, such as plasma protein concentrations and clinical data, in order to obtain systems-level understanding of human immune systems in health and disease.

ABSTRACTS AND SPEAKERS

Unraveling Human Microglial Heterogeneity by Multiplexed Mass Cytometry



Chotima Böttcher, PhD
Senior Scientist and Principle Investigator
Laboratory of Molecular Psychiatry
Charité, University Medicine Berlin

Microglia, specialized innate immune cells of the central nervous system, play crucial roles in brain development and function. In this study we developed a novel cryopreservation protocol and multiplexing technique, which facilitated the comprehensive characterization of small samples (10^3 – 10^4 total cells) of human postmortem microglia (huMG). Using mass cytometry, a core immunophenotypic signature of huMG was identified as compared to cells from peripheral blood and cerebral spinal fluid. We also detected significant regional heterogeneity of huMG using a Cytobank based data processing workflow developed in-house for multidimensional cluster analysis.

Mass Cytometry for Immune Profiling in Chronic Inflammation



Henrik Mei, PhD
Scientific Head of Mass Cytometry
DRFZ Berlin, a Leibniz Institute

We employ high-dimensional mass cytometry to study the immune pathogenesis of chronic inflammation and autoimmunity, and to define disease-specific immune cell signatures that may help guide therapy decisions in the future. I will discuss results of a systematic comparison between blood cells of rheumatoid arthritis subjects and matched controls and will outline several developments that help to generate high-quality mass cytometry data. These include dual fluorescent/metal-tagged antibodies that permit direct cross-platform comparisons, the pre-enrichment of rare cell subsets for downstream analyses by mass cytometry, the use of silver nanoparticles for sensitive detection of weakly expressed antigens and the stabilization of cocktails of metal-labeled antibodies, suitable for longitudinal or multicenter immune phenotyping studies.

ABSTRACTS AND SPEAKERS

Advancing Human Immune Monitoring with an Innovative Panel Kit and Data Analysis Approach



Greg Stelzer, PhD

Director, Mass Cytometry Market Development

Fluidigm

I will present an overview of a new product for use on Helios™ mass cytometry systems, the Maxpar® Human Immune Monitoring Panel Kit. In addition to providing reagents and pre-titrated antibodies that streamline panel optimization and provide reliable cell staining, the kit comes with access to a third-party analysis software solution providing automated population identification and enumeration. I will also present data showing repeatability, reproducibility and performance verification when the panel kit is used with frozen PBMC samples.

New Protocols for Sample Preparation and Acquisition



Dmitry Bandura, PhD

General Manager and Senior Vice President

Canadian Operations

Fluidigm

Fluidigm is committed to empowering the mass cytometry community to achieve new breakthroughs in health and disease. Supporting this goal, we are releasing new cell staining protocols for routine use in mass cytometry to improve cell integrity and staining quality. In addition, we are offering a new sample acquisition protocol for Helios instruments that has been shown to improve data quality. This presentation will include overviews of these new protocols including data supporting their implementation.

ABSTRACTS AND SPEAKERS

Preparing for Clinical Trials in Bergen: From Sample Collection to Data Analysis



Sonia Gavasso, PhD

Researcher, Neuroimmunology Lab and Norwegian
Multiple Sclerosis Competence Center
Haukeland University Hospital and University of Bergen

Mass cytometry has the potential to become a valuable clinical platform to further mechanistic understanding of pathologies and guide therapies. Clinical trials involving this technology offer unique opportunities to link complex data to relevant clinical questions. By optimizing the experimental setup and data analysis pipelines, we are seeking detailed medical answers with the goal of influencing clinical practice in the future. I will discuss, based on our own experiences, the importance of implementing rigorous standard operating procedures for biobanking, the value of pilots and the relevance of building trust across disciplines.

CD Maps: Tracking Antigen Expression During Human Thymic Development



Tomáš Kalina, MD, PhD

Associated Professor, Department of Pediatric
Hematology and Oncology
Charles University in Prague

We attempted to fill in gaps in the known expression of the CD markers across thymocyte development. We developed a fast and accurate time and paths inference algorithm, which involves branching and accounts for possible alternative developmental paths. We have measured human thymocytes using a 37-marker mass cytometry panel (29 surface markers and 8 transcription factors). After exclusion of non-thymocytes (CD56, CD13, CD16, CD33 and CD19-negative), we expert-gated 9 thymocyte stages based on canonical markers (CD34, CD1a, CD3, CD4 and CD8). In parallel, we processed the thymocytes through a computational algorithm. The simulated paths corresponded accurately to the theoretical stages of development and reached the canonical helpers end in 79% and the canonical cytotoxic end in 19%, respectively.

ABSTRACTS AND SPEAKERS

Powering Multi-Omic Translational Studies with Mass Cytometry



Nima Aghaeepour, PhD
Assistant Professor
Stanford University School of Medicine

Recent technological advances in science provide novel opportunities to unravel the complex biology of pregnancy. Immunological changes during pregnancy are highly dynamic and involve multiple interconnected biological systems. An ongoing cohort study by the March of Dimes Prematurity Research Center at Stanford University exploits recent technological advances to examine the transcriptomic, microbiomic and proteomic events associated with normal and pathological pregnancies. I will discuss a machine learning algorithm that will integrate mass cytometry data into this multi-omics setting. This computational pipeline can increase predictive power and reveal new biology by combining datasets of various sizes and modularities in a balanced manner. We expect this approach to be applicable to a wide range of studies beyond the field of pregnancy.

Multidimensional View of the Innate Immune System in HIV-1-Infected Individuals



Antonio Cosma, PhD
Director, FlowCyTech Core Facility
CEA, Université Paris Sud

Human immunodeficiency virus (HIV) infects CD4 T cells, macrophages and monocytes, causing a general dysregulation of the immune system. To obtain a global vision of the modifications induced by HIV-1 infection, mass cytometry and multidimensional data analysis were applied to the study of a group of infected individuals. We applied an analysis pipeline linking event and dimension reduction tools and combining a visualization tool normally used in the business intelligence field.

ABSTRACTS AND SPEAKERS

Optimizing Computer-Assisted Analysis of Immune Response to High-Fat Diet in Mouse Adipose Tissue and Liver



Olivier Molendi-Coste, PhD

Leader, Immune and Metabolism Cytometry Plateau
INSERM, Institut Pasteur de Lille

Obesity and insulin resistance are associated with low-grade inflammation. However, unraveling the global immune adaptations to obesity in a given organ was technically challenging until the recent development of mass cytometry. Using 26 extracellular and 13 intracellular markers, we identified almost all immune cells in epididymal adipose tissue and liver under chow and high-fat diets in mice. We optimized computer-assisted, unbiased analysis by comparing several dimension reduction and clustering algorithms. Finally, viSNE and ClusterX analysis refined the identification of specific recruited and/or activated clusters of effector cells. Application of such an analytic pipeline will provide deeper understanding of cellular local adaptations related to specific pathophysiological and/or therapeutic situations.

Empowering an Imaging Revolution in Tissue Pathology



Minalini Lakshman, PhD

Director of Product Management
Imaging Mass Cytometry
Fluidigm

Imaging Mass Cytometry (IMC™) enables highly multiplexed immunohistochemistry of FFPE and frozen tissues or fixed cells using proven CyTOF® technology. Expanding beyond the limitations of traditional fluorescence-based detection, IMC enables identification of new protein biomarkers and deep profiling of the tissue microenvironment while preserving tissue architecture and cellular morphology. With the launch of the Hyperion™ Imaging System in October 2017, scientists across the world are now empowered to apply IMC in their translational research programs. In my presentation, I will share product updates and successes of the Hyperion Imaging System.

ABSTRACTS AND SPEAKERS

Analysis of Tissue Ecosystems in Health and Disease by Imaging Mass Cytometry



Bernd Bodenmiller, PhD
Assistant Professor
Institute of Molecular Life Sciences
University of Zurich

Human diseases typically occur in tissues and are caused by internal or external factors. Comprehensive analysis and visualization of the tissue ecosystem, including its cell phenotypes, the cellular functions and the cellular interplay, is essential to enable an understanding of tissue biology, to define biomarkers and to identify novel therapies. To enable such comprehensive analysis of tissue ecosystems we

developed Imaging Mass Cytometry (IMC), which enables researchers to visualize >50 markers simultaneously on tissues with subcellular resolution. To fully exploit the potential of IMC, we have developed upstream and downstream experimental and computational workflows that enable co-visualization of transcripts and the generation of mass tomographs. We then applied IMC for the analysis of diseased tissues, including breast cancer samples. Our analyses reveal a surprising level of inter- and intratumor heterogeneity and identify cell-to-cell interaction motifs correlating with clinical outcomes of the analyzed patients. Our results highlight the potential of IMC to provide high-dimensional analysis of cell type, cell state and cell-to-cell interactions in tissue ecosystems. We envision that IMC will become a cornerstone to diagnose a wide range of diseases and to guide treatment in precision medicine approaches in the future.

ABSTRACTS AND SPEAKERS

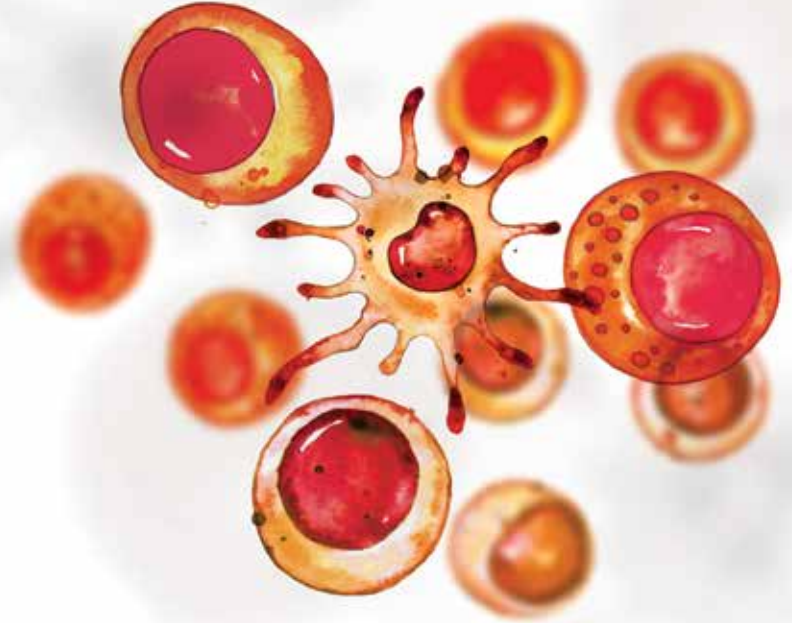
Deep Phenotyping of Colorectal Cancer by High-Dimensional Mass Cytometry Reveals Tumor-Specific Immune Landscapes



Frits Koning, PhD

Professor, Department of Immunohematology
and Blood Transfusion
Leiden University Medical Center

High-dimensional immunophenotyping of colorectal cancers reveals tumor-specific immune signatures and charts the complexity of innate and adaptive immune cell populations in the tumor microenvironment. Previously unappreciated immune cell subsets further differentiate the two main pathways of tumorigenesis in this cancer type. These results may pave the way for optimized and personalized therapeutic approaches.



PANEL DISCUSSION

Slice or Dice? When to Image and When to Suspend Samples for CyTOF Analysis



Moderator

Frits Koning, PhD

Professor, Department of Immunohematology and Blood Transfusion
Leiden University Medical Center



Panelist

Bernd Bodenmiller, PhD

Assistant Professor, Institute of Molecular Life Sciences
University of Zurich



Panelist

Jared Burks, PhD

Assistant Professor, Department of Leukemia
Co-Director, Flow Cytometry and Cell Imaging Core Facility
University of Texas MD Anderson Cancer Center



Panelist

Susanne Heck, PhD

Head of Flow Cytometry Research Platform
NIHR Biomedical Research Centre
Guy's and St Thomas' NHS Foundation Trust and King's College London



Panelist

Jonathan M. Irish, PhD

Assistant Professor of Cell and Developmental Biology
Scientific Director, Cancer and Immunology Core and Mass Cytometry Center of Excellence
Vanderbilt University

TUTORIALS

Join Us at the CYTO® Fluidigm Commercial Tutorials

Establishing Imaging Mass Cytometry in a Core Facility
to Empower Highly Multiplexed Tissue Imaging

Monday, April 30 12:30–1:30 pm South Hall 2B
BOX LUNCH PROVIDED. SEATING IS LIMITED.

Speakers



Susanne Heck, PhD

Head of Flow Cytometry Research Platform
NIHR Biomedical Research Centre
*Guy's and St Thomas' NHS Foundation
Trust and King's College London*



Jared Burks, PhD

Assistant Professor, Department of Leukemia
Co-Director, Flow Cytometry and Cellular
Imaging Core Facility
MD Anderson Cancer Center

TUTORIALS

Mass Cytometry for Clinical Research: Establishing a Systems Immune Monitoring Program

Tuesday, May 1 12:30–1:30 pm South Hall 2B
BOX LUNCH PROVIDED. SEATING IS LIMITED.

Speakers



Jonathan M. Irish, PhD

Assistant Professor of Cell and
Developmental Biology

Scientific Director, Cancer and Immunology Core
and Mass Cytometry Center of Excellence

Vanderbilt University



Greg Stelzer, PhD

Director of Mass Cytometry
Market Development

Fluidigm

FLUIDIGM.COM

Corporate Headquarters

7000 Shoreline Court, Suite 100
South San Francisco, CA 94080 USA

Toll-free: 866 359 4354 in the US and Canada

Fax: 650 871 7152

Sales

North America | +1 650 266 6170 | info-us@fluidigm.com

Europe/EMEA | +33 1 60 92 42 40 | info-europe@fluidigm.com

China (excluding Hong Kong) | +86 21 3255 8368 | info-china@fluidigm.com

Japan | +81 3 3622 2150 | info-japan@fluidigm.com

All other Asian countries | +1 650 266 6170 | info-asia@fluidigm.com

Latin America | +1 650 266 6170 | info-latinamerica@fluidigm.com

For Research Use Only. Not for use in diagnostic procedures.

©2018 Fluidigm Corporation. All rights reserved. Fluidigm, the Fluidigm logo, CyTOF, Helios, Hyperion, Imaging Mass Cytometry, IMC and Maxpar are trademarks and/or registered trademarks of Fluidigm Corporation in the United States and/or other countries. April 2018.