MEETING AGENDA*

Event will take place in Central European Time (CET)

February 24th, 2021

8:00 AM OPENING MESSAGE

DR. AKIRA WATANABE | An application of single cell transcriptomics to regenerative medicine

DR. KOICHI TAKAHASHI | Accelerating life sciences by robotic biology

DR. SAM ABRAHAM, BSC, MSC, PH.D. | High throughput robotics and genomics for an antimicrobial resistance surveillance system for diverse applications

DR. BJÖRN MAGNUSSON, PH.D. | Implementing automation in a next generation sequencing lab to increase capacity and quality in sample processing

DR. JOCHEN HECHT | 16S metagenome sequencing - pitfalls and challenges

LISA TRUONG, PH.D. | Improving high throughput screening to identify chemical bioactivity in zebrafish

MAIKE SEIFERT | From high-throughput sequencing to high-throughput COVID diagnostics

DR. MED. ERIC PAUL BENNETT | INDEL detection, the “Achilles heel” of precise genome editing

MOLLY ZELLER, MS. | Choosing the right DNA-Seq kit for your application

PROF. JOHN W.A. ROSSEN, PH.D. | Decoding the unknown: use of clinical metagenomics to effectively diagnose infectious diseases

DR. CHRISTOPHER E. MASON, PH.D. | High-throughput clinical and environmental viral extraction and sequencing

JASON W. ARNOLD | The pleiotropic effects of prebiotic galacto-oligosaccharides on the aging gut

DR. STEVEN A. SHEARDOWN, PH.D. | Advancing therapeutics against novel targets for CNS diseases using the NETSseq platform

PROF. MARK ADAMS, PH.D. | Genome-based targeted sequencing as a reproducible microbial community profiling assay

DR. JAN HELLEMANS, PH.D. | Applications of qPCR and dPCR in clinical research

DR. TODD DRULEY, MD, PH.D. | Personalized Cancer Monitoring for early and mid-stage solid tumors

B. MELINA CIMLER, PH.D. | Regulatory Insights for Biomarker Development and Clinical Translation

8:35 PM CLOSING REMARKS

9:15 PM END OF THE SYMPOSIUM

*Stay posted for more speakers to come.
An application of single cell transcriptomics to regenerative medicine.

Heterogeneity of gene expression and rarity of replication hampers molecular analysis of β-cell mass restoration in the adult pancreas. Single-cell RNA sequencing of murine pancreases was conducted, with or without partial pancreatectomy, to show transcriptional dynamics in the β-cell replication process. The transcriptional trait describes heterogeneity of Ins1-expressing β-cells, one cluster of replicating β-cells with high expression of cell proliferation markers Pcna and Mki67 was observed. Transient activation of ER stress responders (like Atf6 and Hspa5) was also seen during the transition to replication, along with elevated expression of tumor suppressors – such as Trp53, Rb1 and Brca1 – and DNA damage responders, like Atm, Atr, Rad51, Chek1 and Chek2. This shows the fine balance of cell cycle progression and protection from DNA damage. Taken together, these results provide a high resolution map depicting a sophisticated genetic circuit for replication of the β-cells.

Akira Watanabe is a Program-specific Associate Professor at Graduate School of Medicine, Kyoto University, studying oncology and developmental biology by genomic and epigenomic analysis, including single cell transcriptome analysis.

He graduated from Faculty of Engineering, Tokyo University of Science, and Graduate School of Engineering, the University of Tokyo. He received his PhD in engineering from the University of Tokyo in 2003, and worked as a postdoctoral fellow at the Research Center for Advanced Science and Technology at the University of Tokyo.

He joined the faculty of Center for iPS Cell Research and Application (CiRA) at Kyoto University in 2009, and lead Genomics Core of CiRA.
Dr Koichi Takahashi is a principal investigator at the RIKEN Center for Biosystems Dynamics Research. His research interests span the interface between computing and biology, with the ultimate goal of automating scientific discovery. He is also a Project Professor at Keio University, an Invited Professor at Osaka University, a co-founder and scientific advisor at Epistra Inc, and a technical advisor at Molcure Inc.

Laboratory automation is key to solving problems in today’s life sciences, including poor reproducibility, inefficient operation of expensive laboratory equipment, poorly conducted research and labor-intensive working styles. Although laboratory automation itself is not new, most commercially available products still require human operators who work as the ‘glue’ between machines. In addition to these productivity issues, evident in biological laboratory procedures is a breadth of tacit knowledge, forming a bottleneck in knowledge transfer and reproducibility. The JST MIRAI Project has been developing a formal experimental protocol description language and IoT system architectures to enable coordinated operation of various robots and machinery. It is also creating an AI platform for automated experimental planning and optimization, using data such as cell images to transform biological laboratories into cyber-physical systems. The center has developed an autonomous cell culture system based on LabDroid, a versatile humanoid experimental robot, and succeeded in culture and differentiation of induced pluripotent stem cells into retinal pigment epithelial cells.

Accelerating life sciences by robotic biology.
Antimicrobial resistance (AMR) is one of the most prominent biosecurity issues affecting animals and humans in modern society. AMR in animals is a major global issue in both disease-causing zoonotic pathogens and commensals in the microbiota of healthy livestock. Globally, antimicrobial resistance surveillance and monitoring is widely acknowledged as a critical response to AMR, and is one of the priorities of the WHO Global Action Plan. There are a number of barriers to effective AMR surveillance in food producing animals. In addition, large national surveys undertaken to date are not truly representative of herd level data, and do not provide insights useful for veterinarians and farmers implementing farm control measures via antimicrobial stewardship and infection control. Addressing these barriers requires an inexpensive and accurate means for objectively defining AMR risks at the herd and national level. With the recent development of automated robotic systems and high throughput next generation sequencing platforms, it is now feasible to develop cost-effective tools to monitor AMR on large numbers of representative samples obtained from livestock. Application of this tool to individual herds would deliver an accurate description of their AMR status. This presentation will focus on advancing AMR surveillance through robotic multi-platform integration, and identify balancing the role of phenotype and genotype from a One Health perspective.
Dr Björn Magnusson works in the Transcriptomics and NGS team in the Department of Translational Genomics at AstraZeneca (Gothenburg, Sweden), leading the implementation of automation in the team’s workflows. He has a background in lipid biology and metabolic disease from his PhD and post-doctoral research at Gothenburg University. In 2010, he left academia to work on in vitro assays in the Diabetes Research Unit at Novo Nordisk in Copenhagen, Denmark. Björn joined the cell assay development team at AstraZeneca in 2011, and moved into next generation sequencing in 2015. He has used automated liquid handling platforms of various types throughout his work at AstraZeneca and, since 2018, has been the superuser of the department’s Fluent Automation Workstation.

The NGS Transcriptomics team at AstraZeneca partners with scientists across AstraZeneca and externally, to enable transcriptomic analysis in various projects across our therapy areas. Björn will present how the team applies automation for its work, describing how they developed from low throughput, all manual work to utilizing automated liquid handling for their core processes. These include preparation of high quality RNA from various samples, sample normalization and randomization, library preparation and pooling of samples. Particular focus will be given to protocols for automated NGS library preparation, which have been established on Fluent 1080 system. These are full-day protocols that can process 8-96 samples through multiple enzymatic reactions, on-deck PCR runs, bead-based purifications, etc. He will also discuss the impact of automation on the team’s work in terms of quality and capacity, exemplifying with some recent projects.
Jochen undertook did his PhD in Stefan Mundlos’ group at the Max Planck Institute for Molecular Genetics in Berlin, Germany. In 2008, he set up the first NGS unit at the Charité University Hospital Berlin, before moving to Barcelona in 2015 to lead the Genomics Unit of the Centre for Genomic Regulation. As a service provider, the CRG Genomics Unit does not perform independent research, but is fully focusing on supporting researchers by performing small to large scale NGS experiments of all types.

16S metagenome sequencing - pitfalls and challenges.

Metagenomics analyses has become increasingly popular across many fields of research in the past few years. For profiling bacterial communities, the 16S rRNA gene has become one of the most frequently used methods, due to its ease of use and low costs per sample. This presentation aims at highlighting some of the pitfalls and challenges related to 16S library preparation and sequencing.
Lisa Truong is currently an Assistant Professor in the Department of Environmental and Molecular Toxicology, and the Deputy Director of the Sinnhuber Aquatic Research Laboratory at Oregon State University, where her research program goal is to utilize the zebrafish model to build computational models to be less reliant on animal testing and conduct toxicity-testing based on toxicity pathways. She received her Ph.D. in Toxicology at Oregon State in 2012 and was a postdoctoral fellow at US EPA-National Center for Computational Toxicology. Lisa has been conducting research utilizing the zebrafish model for the last 15 years.

There is limited safety information available for most of the chemicals humans are exposed to. To aid in filling this data gap, we need efficient screening methods to estimate the chemicals hazard potential. The ideal model is the embryonic zebrafish. Zebrafish embryos develop rapidly, share 80% gene homology with higher vertebrates, and due to their size, highly amendable to high throughput automation. The zebrafish assays can detect bioactivity at the molecular, phenotypic, and behavioral levels. By adopting new technologies, the efficiency and accuracy of bioactivity identification have improved dramatically and has moved the zebrafish model to be the premier bioactivity screening model for human health hazards.
Maike received her BSc in cell biology from the University of Osnabrueck (Germany), and a master’s degree in molecular and applied botany from the University of Hamburg, and conducted three years of PhD studies at the Max Planck Institute for Molecular Plant Physiology in Golm.

In 2015, she moved to Stockholm and joined Lars Engstrand’s group at the Karolinska Institute, first in Clinical Genomics, then later at the Center for Translational Microbiome Research, where she has developed and optimised an mostly automated, high throughput microbial amplicon and metagenome pipeline.

2020 was a special year for all of us. At the end of March 2020, the Center for Translational Microbiome Research was quickly converted from a large-scale microbiome research lab into a SARS-CoV-2 testing center, to assist in analyzing SARS-CoV-2 PCR tests from across Sweden.

Initially assisting with RNA extraction for the National Pandemic Center at the Karolinska Institute/SciLifeLab, the Center for Translational Microbiome Research rapidly expanded its involvement, becoming one of the biggest testing facilities in Sweden, with a testing capacity of 10,000 samples per day, typically turning around results within 24 hours from samples arriving at the lab. This transformation would not have been possible or sustainable without Tecan.
INDEL detection, the “Achilles heel” of precise genome editing.

Advances in genome editing technologies have enabled manipulation of genomes at the single base level. These technologies are based on programmable nucleases (PNs) that include meganucleases, ZFNs, TALENs and CRISPR/Cas9, and have enabled researchers to delete, insert or replace genomic DNA in cells, tissues and whole organisms. The great flexibility in redesigning the genomic target specificity of PNs has vastly expanded the scope of gene editing applications in life sciences, and shows great promise for development of the next generation gene therapies. PN technologies share the common principle of inducing a DNA double-strand break (DSB), followed by cellular repair of the induced DSB, resulting in the formation of insertions or deletions (InDels) at the user-specified genomic site. This presentation outlines how methods have been developed for detection of the essential PN-induced primary outcomes, and how recent advances enable turning of a complex pool of different InDel events into informative InDel profiles.
MOLLY ZELLER, MS.
University of Wisconsin, USA

Choosing the right DNA-Seq kit for your application.

The DNA Sequencing Core Facility at the University of Wisconsin–Madison Biotechnology Center (UWBC) has extensive experience generating a multitude of next generation sequencing (NGS) libraries. The facility works with various sample types and species— including humans, mice, microbiomes, fungi, and many more—from all over the world. With so many libraries to prepare, the team became increasingly aware that the library preparation kit that they were using was laborious, expensive and, most importantly, provided inconsistent results. There were a number of reasons that they believed were contributing to this poor kit performance, and so the team began looking for a kit offering a quick, budget-friendly and uniform workflow that addressed these issues. In this talk, Molly will explain why her group chose the Celero™ EZ DNA-Seq Library Preparation Kit for its genomic DNA library preparation workflow, and how this kit performed against other DNA-Seq library prep kits on the market.
John Rossen has a 30-year history in molecular virology and microbiology and is Professor for Medical Microbiology - in particular Personalised Microbiology - at the University of Groningen. His Personalised Microbiology group has implemented NGS into clinical microbiology and infection prevention, and is now focusing on implementing metagenomics and metatranscriptomics. These methods are applied to patient, animal, food and water samples, characterizing microorganisms (including viruses) and the interaction between them, as well as with their host. He is immediate past president of the ESCMID study group for genomic and molecular diagnostics (ESGMD), and board member of the Dutch Society of Medical Microbiology.

University of Groningen, Netherlands / IDbyDNA, USA

Decoding the unknown: use of clinical metagenomics to effectively diagnose infectious diseases.

Metagenomics uses direct sequencing of clinical specimens without culturing, and bioinformatics techniques to identify and characterize microorganisms, including DNA/RNA viruses, bacteria, fungi, protozoans, and helminth viruses and bacteria. By determining the entire genomic sequence of a pathogen, scientists and clinicians can decipher not just the organism, but also strain type, which informs transmission patterns and identifies sequences known to be antimicrobial resistance (AMR) markers. AMR markers inform treatment options by indicating which antivirals or antibiotics will not be effective against the target pathogen. Moreover, metagenomics allows the detection of co-infections that, eg. can be the cause of death in many COVID-19 patients. In this presentation, the implementation of metagenomics in clinical microbiology will be discussed, and examples of its use in real life will be presented.
Dr. Christopher Mason is an Associate Professor of Genomics, Physiology and Biophysics at Weill Cornell Medicine, and the Director of the WorldQuant Initiative for Quantitative Prediction. He also holds appointments at the Tri-I Program on Computational Biology and Medicine (Cornell, Memorial Sloan Kettering Cancer Center and Rockefeller University), Harvard Medical School, and Yale Law School. The Mason laboratory develops and deploys new biochemical and computational methods in functional genomics to elucidate the genetic basis of human disease and physiology. They create and deploy novel techniques in next generation sequencing, and algorithms for tumor evolution, genome evolution, DNA and RNA modifications, and genome/epigenome engineering. They also work closely with NIST/FDA to build international standards for these methods (SEQC2, IMMSA, and Epigenomics QC groups), to ensure clinical quality genome measurements and editing. They also work with NASA to build integrated molecular portraits of genomes, epigenomes, transcriptomes and metagenomes for astronauts, which help establish the molecular foundations and genetic defenses for enabling long-term human spaceflight.

COVID-19 is a severe disease that has caused >1 million deaths in under one year. As this disease is novel, the molecular and cellular underpinnings of the progressive tissue injury are poorly understood, as is the role of direct versus indirect viral-induced injury. This study compared spatially-resolved gene expression in fixed lung tissue from autopsies of COVID-19 patients with high and low viral loads to those who suffered from other respiratory diseases – flu or non-viral acute respiratory distress syndrome (ARDS) – and normal lung tissue. We used the DreamPrep™ NAP workstation to extract RNA and DNA to profile 735 COVID-19 patients. Using normal samples as a reference, there was an enrichment of genes involved in antigen presentation and interferon response in high and low viral load patients. Between COVID-19 cohorts, there was an overall upregulation of individual interferon-stimulating genes in COVID-high patients, and an increase in collagen genes found in COVID-low patients. Pathway analysis revealed enrichment in interferon alpha/beta and gamma signaling in COVID-19 samples compared to normal, and this enrichment scaled with viral load. COVID-high patients showed enrichment in these pathways compared to flu and ARDS patients, while COVID-low patients had enrichment of collagen-related pathways compared to flu and ARDS.
The pleiotropic effects of prebiotic galacto-oligosaccharides on the aging gut.

Dietary prebiotics - indigestible carbohydrates that are selectively fermented by gut micro-organisms - are emerging as safe and effective modulators of the gut microbiota. Prebiotic galacto-oligosaccharides (GOS) are now included in infant formula, due to their similarity to human milk oligosaccharides, and benefit microbiota modulation during early development. As we age, the composition and function of the gut microbiota changes. A dysbiotic microbiota associated with aging contributes to physiological aberrancies in the gastrointestinal tract. The goal of this study was to assess the impacts of dietary GOS on the aging microbiota and on gastrointestinal physiology. By using an aging mouse model, the team performed a longitudinal dietary study assessing factors including intestinal permeability, inflammation, gene expression, microbiota composition and microbiota functional capacity. Results show that dietary GOS increased the abundance of beneficial micro-organisms, increasing the saccharolytic potential of the microbial community, improving intestinal barrier, increasing mucus production and modulating gene expression.
Dr Steven Sheardown received his BSc in Genetics from Sheffield University, and his PhD in Somatic Cell Genetics from Edinburgh University.

He went on to postdoctoral research with the Medical Research Council, using mouse knockout to generate disease models and, subsequently, researching regulation of the mouse Xist gene. From there, he moved to GSK as a founding member of the Transgenic Technologies Group, followed by taking a position with Takeda Cambridge as Transgenics Group Head.

Steven joined Cerevance in 2016 to head Molecular Biology, with a remit to transform NETSseq into a high throughput CNS target discovery platform.

Cerevance is a private pharmaceutical company focused on central nervous system diseases. Its proprietary NETSseq technology enables the company to produce comprehensive transcriptional profiles of specific brain cell types from mature human brain tissue. Steven will describe the rationale for this approach, how the NETSseq method works, and how Cerevance has used the Trio RNA-Seq Library Preparation Kit to generate good data from difficult materials. To date, the company has produced over 5,000 high quality, cell type-specific datasets from control and disease tissues, and is well positioned to deliver life-changing therapeutics for patients who have brain-related disorders.
Dr Mark Adams is Professor and Deputy Director at The Jackson Laboratory for Genomic Medicine (JAX), where he leads the Clinical Diagnostic Research group.

Dr Adams was a co-founder of The Institute for Genomic Research and Celera Genomics. Prior to joining JAX, he was Associate Professor of Genetics at Case Western Reserve University and then Scientific Director at the J. Craig Venter Institute.

Dr Adams has a BA in Chemistry from Warren Wilson College, and a PhD in Biological Chemistry from the University of Michigan.

Genome-based targeted sequencing as a reproducible microbial community profiling assay.

Current sequencing-based methods for profiling microbial communities rely on marker gene (e.g. 16S rRNA) or metagenome shotgun sequencing (mWGS) analysis. Mark will present an approach based on a single-primer extension reaction, using a highly multiplexed oligonucleotide probe pool based on Allegro Targeted Genotyping. This approach – termed MA GenTA (Microbial Abundances from Genome Tagged Analysis) – enables quantitative, straightforward, cost-effective microbiome analysis. The use of multiple probes per target genome, and rigorous probe design criteria, enabled robust determination of relative abundance. Probes were designed for 830 genome sequences representing bacteria present in mouse stool specimens. Comparison of the MA-GenTA data with mWGS data demonstrated excellent correlation down to 0.01% relative abundance. Despite the incompleteness of the reference database, NMDS clustering of experimental sample groups was consistent between MA-GenTA, mWGS and 16S rRNA datasets. MA-GenTA represents a potentially useful new method for microbiome community profiling based on reference genomes.
Jan Hellemans is founder and CTO of Biogazelle, a Belgian service provider exploiting RNA to improve healthcare. He is an expert in qPCR and dPCR technologies, and co-author of the Digital MIQE Guidelines. He led the development of Bio-Rad’s PrimePCR assays, and supports Biogazelle’s service team in designing and validating qPCR/ddPCR assays for clinical research.

Applications of qPCR and dPCR in clinical research.

Quantitative PCR (qPCR) and digital PCR (dPCR) are cornerstone technologies in most, if not all, molecular labs. They are used for quantification of certain sequences, be it the expression of a gene, the copy number of a transgene or the viral load. While dPCR enables nucleic acid quantification with superior accuracy and precision compared to qPCR, it also comes with drawbacks, such as increased costs and lower dynamic range and throughput. A thorough understanding of the limits and advantages of these 2 PCR variants is essential in choosing the right application for the research question being faced, especially in the context of a clinical trial with tight regulatory constraints.

This talk will describe the intrinsic potential of qPCR and dPCR in clinical research, presenting case studies in the space of SARS-CoV-2 detection and CAR-T studies. For each case study we will elaborate on the rationale for selection of qPCR vs dPCR.
Dr Todd Druley is the Chief Medical Officer for Somatic Oncology at Invitae. He obtained a BSc in cell and structural biology, and a minor in chemistry, from the University of Illinois in 1994. He then completed the MD/PhD program at the University of Illinois, where he studied mechanisms of chemotherapy resistance. In 2002, Todd joined Washington University as a pediatric resident and has since completed his fellowship in pediatric hematology and oncology, joining the faculty in 2008. Research in the Druley Lab is based on improving molecular diagnostics in pediatric AML. He is also a member of the Children’s Oncology Group (COG) Myeloid Disease and Epidemiology Committees, and a board-certified pediatric hematologist/oncologist and Associate Professor of pediatrics, developmental biology and genetics at Washington University School of Medicine.

Invitae’s Personalized Cancer Monitoring (PCMTM) research panel, built with Anchored Multiplex PCR (AMPTM) chemistry to detect molecular residual disease (MRD), is a bespoke, tumor-informed liquid biopsy panel designed to provide best-in-class sensitivity and specificity to identify residual or recurrent cancer earlier than the current standard of care. The TRACERx study, conducted in collaboration with Cancer Research UK, demonstrated 93% of patients with relapsed NSCLC were detected prior to current methods with a median lead time of 136 days (maximum 1,022d), nearly twice the lead time previously reported, and a limit of detection of 0.003% allele frequency. PCM demonstrated 99.3% specificity, suggesting capability for more frequent testing than current MRD assays. In summary, the precision offered by Invitae’s PCM panel may enable MRD monitoring as a surrogate clinical trial endpoint, resulting in faster, less-expensive studies, and earlier detection of recurrent cancer facilitating rapid salvage therapy intended to improve patient outcomes.
Dr Melina Cimler is the CEO and Founder of PandiaDx LLC. She has over 30 years of experience in the life sciences and diagnostics industry, leading regulatory, quality, clinical affairs, research and product development organizations with a focus on precision medicine. She served as Senior Vice President of Regulatory & Quality at Adaptive Biotechnologies until April 2018, and was previously Head of Global Quality and Vice President of Quality, Regulatory, Clinical and Government Affairs at Illumina. She defined and executed the regulatory strategy for FDA marketing authorization of the MiSeqDx NGS platform. Melina also serves on the Board of Directors of Nanostics, as a member of the Scientific Advisory Board of Athira Pharma, and as an Expert Consultant at NDA Partners. She holds a Ph.D. in Pharmacology from the University of Washington.

Genomic advances are driving more precise therapies that are rapidly evolving. To enable the precision medicine revolution, biomarker development, validation and translation into clinical practice require careful navigation of regulatory pathways. Expectations for tumor profiling versus companion diagnostics are different. With focus on COVID-19 marketing authorizations, regulators have limited resources for supporting review of other products. Successfully choosing and navigating the appropriate pathways will determine market access.