USP/BioPhorum Joint Workshop

ADRESSING BARRIERS to ADOPTION

Abstracts and Speaker Profiles

Welcome!

On behalf of US Pharmacopeia, BioPhorum and the workshop organizing committee, we would like to welcome you to the **2022 Joint Workshop on Continuous Manufacturing of Biologics: Addressing Barriers to Adoption***. We have gathered an exciting slate of speakers, and over the next two days you will hear talks on various aspects of continuous manufacturing of biologics. We have left plenty of time for questions and panel discussions after each session, so please have your questions and comments ready. There will also be a reception after the first day, for in-person attendees, where we will have a chance to meet and exchange ideas in a less formal setting. Thank you for attending, and we look forward to an exciting workshop.*

Organizing Committee

Rich Chen, *Exec Dir Purification & Viral Safety, Eli Lilly and Company*

Chris Hwang, *Chief Technology Officer, Transcenta Therapeutics Inc.*

Maura Kibbey, *Director, Biologics Marketing, US Pharmacopeia*

John Kokai-Kun, *Director, External Scientific Collaboration, US Pharmacopeia*

Julie Kozaili, *Senior Scientist, Asahi Kasei Bioprocess*

Graeme Moody, *Program Manager, BioPhorum*

Kristina Pleitt, *Sr. Manager, Bioproduction R&D Innovation*, *Thermo Fisher Scientific*

Mark Schofield, *Senior R&D Manager, Pall Biotech*

Andrew Zydney*, Bayard D. Kunkle Chair and professor of chemical engineering, Penn State College of Engineering, Pennsylvania State University*

Thank you to USP support staff:

George Athas and Claire Astle for supporting stakeholder outreach, Ginger Fletcher for meeting organization and website support and Peter Suh and Ernest Musser for technical support.

Organizing Committee

Rich Chen, *Exec Dir Purification & Viral Safety, Eli Lilly and Company*

Richard Chen is currently an Exec Director within the Purification Development & Viral Safety Group at Eli Lilly responsible for the development of early/late phase protein therapeutics and Next Generation Bioprocessing initiatives. He has over 30 years of experience in development and manufacturing at various companies including Neose Technologies, Merck, GlaxoSmithkline, Johnson & Johnson (Centocor), Wuxi Apptec and ImClone Systems. He received his BSE degree in Biomedical Engineering from the University of Pennsylvania. He is an active member of the Innovation & Quality (IQ) and the National Institute of Innovation for Manufacturing Biopharmaceuticals (NIIMBL) consortiums.

Chris Hwang, *Chief Technology Officer, Transcenta Therapeutics Inc.*

Chris Hwang has over 30 years of experience in biopharmaceutical industry, mainly in the areas of CMC development, manufacturing, and support. At Transcenta, he is responsible for developing and implementing advance biomanufacturing platform to dramatically lower cost of goods to expand patient access to innovative biologics. Prior to joining Transcenta in 2016, he spent 25 years at Genzyme and Sanofi, most recently as Senior Director in Late-Stage Process Development and was the program lead for the integrated continuous biomanufacturing (ICB) platform. He also has led a number of clinical and commercial stage products at Genzyme and Sanofi. He received his Ph.D. in biochemical engineering from M.I.T.

Maura Kibbey, *Director, Biologics Marketing, US Pharmacopeia*

Dr. Maura Kibbey is Senior Scientific Fellow for Education and Training in USP's Global Biologics Department. Dr. Kibbey collaborates with scientific experts and trainers to bring more educational offerings to USP's biologics stakeholders. Previously, Maura directed a team of liaisons working with the five USP Expert Committees and multiple Expert Panels for biologics, peptides, and antibiotics to develop standards that support biopharmaceutical quality assessment and development. Before joining USP, Dr. Kibbey worked for several biotechnology and diagnostic companies in the Washington DC area as well as at the National Institutes of Health. Her scientific expertise includes development and validation of many different assay types for measurement of individual molecules, their activities, or binding

interactions. She has published over 40 peer-reviewed articles and has been an invited speaker or workshop organizer for numerous scientific conferences.

John Kokai-Kun, Ph.D., *Director, External Scientific Collaboration, US Pharmacopeia*

John F. Kokai-Kun, PhD is Director, External Scientific Collaboration for the US Pharmacopoeia. Dr. Kokai-Kun received his PhD in Microbiology from the University of Pittsburgh, School of Medicine and has more than 20 years of experience in drug development. He has held various positions with several biotechnology and pharmaceutical companies where his research and development efforts have focused primarily biologics and vaccines. Dr. Kokai-Kun was also an Adjunct Assistant Professor of Microbiology and Immunology at the Uniformed Services University of the Health Sciences and is retired from the United States Army.

Julie Kozaili, *Senior Scientist, Asahi Kasei Bioprocess*

Julie Kozaili is a senior scientist and manager in the Research and Development team, part of the Science and Technology department at Asahi Kasei Bioprocess in Glenview, Illinois. Julie's work focuses on supporting Planova customers through the design and execution of studies aimed at better understanding the capabilities and limitations of the virus filters. As part of this role, Julie maintains several collaborations with industry partners, many focused on continuous bioprocessing, with the goal of improving customers' ability to provide safe biotherapeutics to patients around the world. Prior to joining AKBA, Julie received a Ph.D. in Physiology and Biophysics from the University of Illinois at Chicago and was part of the Process and Analytical Development teams at a pharmaceutical company focused on developing biosimilar products.

Graeme Moody, *Program Manager, BioPhorum*

Graeme is a Program manager for BioPhorum, based in Cambridge UK. He has 25 years in the pharma industry as a drug metabolism and pharmacokinetics specialist, supporting oncology, respiratory/inflammation and diabetes research programmes. Graeme introduced some of the first robotic liquid handlers into Astrazeneca in the mid-1990s. Latterly he is a Lean Sigma Master Blackbelt and has lead numerous business improvement projects within R&D at Astrazeneca. Graeme has been with BioPhorum for 5 years and is running projects on Reducing Product Release Time and Continuous bioprocessing.

Kristina Pleitt, *Sr. Manager, Bioproduction R&D Innovation*, *Thermo Fisher Scientific*

Kristina Pleitt is a Senior Manager in Thermo Fisher Scientific's Bioprocessing Collaboration Center part of the R&D Innovation team. She is responsible for leading cross-Group product and process development projects. The majority of Kristina's industry experience is in downstream process development at a CDMO (Thermo Fisher's Pharma Services Group; Patheon). She has experience with full-development and tech transfers for a variety of early and late Phase recombinant proteins including scale-up, GMP production, process characterization, and process validation. Kristina has a BS in Chemical Engineering and is completing her PhD in Industrial Biotechnology (focused on continuous bioprocessing) at The University of Queensland.

Mark Schofield, *Senior R&D Manager, Pall Biotech*

Mark Schofield earned his degrees in Scotland, he received his bachelor's degree from the University of Edinburgh and his molecular biology Ph.D from the University of Dundee. For the last 11 years he has been an employee of Pall life sciences focusing on chromatography applications. Currently he holds the position of Senior R&D manager, his team works on chromatography solutions for continuous bioprocessing and gene therapy modalities.

Andrew Zydney, *Bayard D. Kunkle Chair and professor of chemical engineering, Penn State College of Engineering, Pennsylvania State University*

Andrew Zydney is the Bayard D. Kunkle Chair and professor of chemical engineering at the Penn State College of Engineering, Pennsylvania State University. He received his BS, Chemical Engineering, Yale University and his Ph D, Chemical Engineering, Massachusetts Institute of Technology. His interest areas include Membrane Separation Systems for Bioprocessing and Medical Devices: Purification of high-value biological products including recombinant proteins, gene therapy agents, and vaccines; application of membrane systems for bioseparations and artificial organs.

DAY 1 – Wednesday, December 7, 2022

USP Welcome

Fouad Atouf, Ph.D.
Vice President, Global Biologics

Fouad Atouf is Vice President, Global Biologics, for USP. He leads all scientific activities related to the development and maintenance of documentary and reference standards for biologics and antibiotics, and oversees the biologics laboratories in USP–U.S. and USP–India. His department supports the work of the associated USP Expert Committees. Dr. Atouf has been at USP for over 10 years and served in a variety of scientific leadership roles including being the regional champion for the Middle East and North Africa Region, where he helped facilitate programs designed to enhance the understanding of the role of regulations and standards in the registration of medicinal products. Dr. Atouf has strong background and experience in the development and regulation of *cellular and tissue-based products*. Prior to joining USP in 2006, his research at the U.S. National Institutes of Health focused on developing methods for the *in vitro* generation of cell-based therapies for diabetes. Dr. Atouf is the author of numerous publications in peer-reviewed journals and a frequent speaker at national and international scientific conferences. Dr. Atouf earned his Master's degree in Biochemistry and his Ph.D. in Cell Biology from the Pierre & Marie Curie University, Paris, France.

Session I – Development and Control Strategies

Session Chair: Mark Schofield, Senior R&D Manager, Pall Biotech

Kurt Boenning
Pall Corporation

Kurt Boenning is an R&D Engineer II for the Pall Corporation. He received his BS in Chemical and Biological Engineering from the University of Colorado. He is currently an Engineer II and has worked at Pall for just over four years on chromatography applications. His main areas of focus have been process development for gene therapies and process intensification.

Title: Automation and Control of an Integrated Continuous Bioprocess
Authors: Kurt Boenning, Terése Joseph, Christina Caporale, and Mark Schofield

Abstract: The process intensification of biomanufacturing is being actively pursued. The promises of a more modern manufacturing process include improved control and consistency along with more efficient production which may enable treatment of new patient populations. Due to the high cost of the protein A capture step, alternative processing modalities, such as multicolumn chromatography, have become of interest to

increase process efficiency and decrease cost. Subsequent interdependent unit operations comprising of low pH virus inactivation, dead end filtration, ion exchange chromatography steps, and tangential flow filtration harmonize to create the desired drug substance. To facilitate such an interconnected and complex sequence of unit operations an automated control platform is needed. The relationship between the automation and process is one where the automation is used to a) mitigate the biggest risks associated with the process, b) repeat procedural practices to minimize human intervention or errors, and c) accumulate and store all relevant process information to feedback into system for control.

A fully automated approach to the control of such a continuous downstream process has been evaluated for its efficacy to regulate a process with minimal intervention from operators. Enacting this control strategy facilitated an end-to-end process for 24 hours at two scales as well as a small scale 4-day process. These processes kept target product quality throughout and were executed with minimal operator intervention. Some particular challenges, such as volumetric throughput limitations, scale-up, equipment sizing, etc. were explored in a lab scale setting up to a 20x scale up for the protein A capture step.

By operating our downstream process within the appropriate design space, a deeper fundamental understanding of the purification platform can be developed. This operation seconds as a proving ground for the control strategy. By operating the process, deficiencies in the control can be identified and optimized to improve the efficiency, efficacy, and product quality. Through these means, further understanding of critical process parameters and how they affect the critical quality attributes at the whole process level, not just at the individual unit operation level, has been achieved. Using the knowledge gained from these studies we can identify which parameters have the largest impact on the process and which should be controlled within the tightest engineering limits from a process perspective and further develop and implement automation improvements to eliminate control deficiencies.

Nate Ostberg
Sanofi

Nate Ostberg is a senior scientist in upstream process development at Sanofi where he has developed early and late-stage processes in fed-batch and perfusion cell culture. He has been active in teams to improve the platform for process development, streamline tech transfer to DS manufacturing, and decrease the time from research to IND and FIH studies. He has a chemical engineering background and appreciates an efficient, robust interface from cell line generation to cell culture and purification development. Nate is especially motivated to bring drugs to market for rare diseases, at Sanofi working on blood factor and enzyme replacement therapies.

Title: Rapid Drug Substance Delivery of a Complex Protein Manufactured with a Perfusion Process

Authors: Amanda Ramsdell, Nathan Ostberg, Yang Wu, Shawn Barrett, Henry Lin, Daryl Powers

Abstract: Efficiency and speed to First in Human trials is a critical development objective for early clinical programs. Here we will present a case study for the rapid advancement of a complex therapeutic protein manufactured with a perfusion process. In this case approximately 35 weeks passed between first transfection and availability of drug substance for toxicology studies, and clinical manufacturing could have been initiated after an additional 3 weeks. Several strategies were employed to enable accelerated development including use of pools for early material generation to support downstream, analytical, and formulation development; scale down perfusion models for clone selection; and use of a perfusion platform to minimize process development activities. Using these approaches, drug substance process development timelines and resource utilization were not significantly increased beyond those for a typical monoclonal antibody fed-batch process.

Lara Fernandez-Cerezo
Merck & Co

Lara Fernandez-Cerezo pursued an engineering industrial PhD collaboration between University College London and Merck focusing on scale-down filtration technologies of high concentration mAbs. Upon graduation, she established the ambr® crossflow high throughput system as a process characterization/development UF/DF tool in the Merck process development teams. In early 2019 she transitioned to continuous manufacturing (CM) in biologics gaining hands-on experience in both non-GMP 50L development scale and GMP 500L clinical scale. Since moving back to the development group in the US in 2021, Lara continues to influence the GMP continuous manufacturing teams at Merck by providing recommendations for the design of Merck's next generation commercial CM facility including leading the design of a customized SPTFF/ILDF Skid. Lara now leads the downstream biologics continuous manufacturing team in the Enabling Technology group at Merck. Other activities she is involved with include co-leading a Merck / Penn State Collaboration focusing on scale-up depth filtration challenges; and leading an IQ sub-group focusing on the use of UF/DF for post-conjugation steps in antibody-drug conjugates (ADCs).

Title: Development Roadmap of Continuous Manufacturing Operations at Merck

Authors: Lara Fernandez Cerezo[1], Nuno Pinto[1], Adrian Gospodarek[1], Will Rayfield[1], Julie Kozaili[2], Daniel Strauss[2] Matt Kessler[3], Mark Brower[1]

1 Merck & Co., Inc, Kenilworth, NJ, USA
2 Asahi Kasei Bioprocess America
3 MSD, Lucerne, Switzerland

Abstract: Standard fed-batch (FB) monoclonal antibody biologics manufacturing processes are costly and time-consuming. A switch to intensified continuous

manufacturing (CM) processing enables much higher volumetric productivity than traditional batch/fed-batch processes with 2 to 5-fold higher cell density and cultivation duration.

Three topics will be covered in this talk; 1) choice of cell retention tangential flow filtration (TFF) perfusion device; 2) end-to-end steady state operation; and 3) impact of low flux and pressure interruptions on viral clearance.

One challenge that remains in cell retentate hollow fiber TFF systems is to further understand the decaying product sieving profile observed during the extended cell culture operations. Maximizing product sieving helps mitigate detrimental impact on protein quality attributes and ultimately improve on overall process yield. A case study will be presented comparing three commercially TFF membranes (with various pore size and chemistries) tested across a range of operating conditions in high-cell density CHO perfusion for 21+ culture days. This work helped identify a filter which offered high mass transfer (>90% sieving) and particle-free permeate stream (turbidity <10 NTU) to allow direct loading of a chromatography capture step.

A main distinguishing factor of CM compared to traditional FB is the end-to-end operation with uninterrupted medium exchange and perfusate flow supply in upstream into fully connected downstream. This is achieved via end-to-end flowrate alignment dependent on the perfusate titer. This replaces the need of large hold intermediate tanks with significantly smaller surge vessel tanks in between unit operations to help manage process pauses and ensure flow continuity (especially due to slow yet continuous flowrates).

This is particularly important for viral clearance, more specifically when evaluating if commercial viral filters can withstand the expected low flux conditions and potential pressure interruptions. A case study will be presented using four different minute virus of mice (MVM)-spiked feeds and small-scale hollow fiber virus filters with hydrophilic modified polyvinylidene fluoride (PVDF) membrane. Low flux, high volumetric throughput, filter fouling and process interruptions showed minimal impact on overall MVM log-reduction virus (LRV) achieving >4 log reduction when running continuously up to 6 days.

Based on small-scale data, decisions can be made to identify best-performing filters and define filter switch criteria for example, based on sieving cut-off (TFF-based perfusion) and target volumetric throughput and number of pauses (VF). All these case studies not only showcase additions to the CM toolbox, but also demonstrate that CM is possible with TFF-based perfusion cell culture and commercially available virus filters and have even been implemented for GMP processes. Unique challenges to CM can be potentially overcome by tailoring the process design helping to ultimately increase operational flexibility.

Session II – Lessons Learned & Case Studies

Session Chair: Julie Kozaili, Senior Scientist, Asahi Kasei Bioprocess

Michael Coolbaugh
Sanofi

Michael joined the purification process development team at Sanofi in 2014 after receiving his PhD in Chemical and Biomolecular Engineering from Ohio State. Since joining Sanofi, Michael has led the development of purification processes and drug substance teams for a variety of molecules in Sanofi's pipeline, ranging from monoclonal antibodies to enzyme replacement therapies. Michael has also been very active in Sanofi's integrated continuous biomanufacturing (ICB) technology development efforts, with an emphasis on extending the continuous boundary to encompass the final downstream process steps. Michael is currently leading Sanofi's end-to-end ICB team from the proof-of-concept phase to GMP implementation and commercialization.

Title: Integrated Continuous Biomanufacturing in GMP Settings: Present State and Future Outlook

Authors: Michael Coolbaugh, Rohan Patil, Kevin Brower, Jason Walther*Associate Director, Purification Process Development Purification Process Development, Global CMC Development, Sanofi*

Abstract: The biopharmaceutical industry has seen significant and exciting advancements in the area of integrated continuous biomanufacturing (ICB) in recent years. Multiple companies and academic institutions have reported innovations in enabling technologies and strategies for continuous manufacturing coming out of their development labs. However, innovation is not only occurring in the lab. Here, we will present on the successful commercialization and regulatory approval for two molecules using our hybrid ICB GMP manufacturing platform, with an emphasis on innovations in process validation and regulatory strategies. We will then further discuss the continued development of our ICB platform and the path forward to the implementation of end-to-end continuous manufacturing (bioreactor to drug substance) in GMP manufacturing.

Charlie Heise
FUJIFILM Diosynth Biotechnologies

Charles Heise is a Senior Staff Scientist in the Bioprocessing Strategy & Development group at FUJIFILM Diosynth Biotechnologies working on developing connected, integrated processes. He has 15 years of experience in leading the development and scale up of manufacturing processes for clinical and commercial stage biologics. He is co-inventor of the award winning SymphonX™ purification skid and the downstream technical lead for FDB's continuous biomanufacturing process (MaruX™).

Title: MaruXTM: Deployment of a Control Strategy for a 500 L Continuous Biomanufacturing Platform

Abstract: Continuous manufacturing provides a solution for uncertainty in capacity as drug candidates progress through clinical development to commercial supply. However, adoption has been slow due to technical and operational challenges associated with managing large liquid volumes, integration of unit operations, and environmental and process controls. We present a case study describing the successful production of a monoclonal antibody using a 500 L scale connected process with a single-use flow path in a non-GMP pilot facility, which is a forerunner to the construction of flexible continuous GMP manufacturing facilities.

Using our proprietary ApolloX™ high-biomass 'perfusion ready' cell line, combined with a commercially available single-use bioreactor operated in perfusion mode linked to our in-house multi-functional SymphonX™ purification systems, with integrated point-of-use buffer dilution, we were able to produce and purify ~1 kg mAb/day from the perfusion reactor over a representative 2 week period. The presentation will outline the regulatory considerations, technical solutions and control strategy deployed to mitigate the complexities of continuous bioprocessing operations in a multi-product facility.

Joel Bruegger, Ph.D.
Parvus Therapeutics

Joel Bruegger is Associate Director of Protein Science and Process Development at Parvus Therapeutics. He has 15 years' experience in protein sciences that includes earning a Ph.D. in Biological Sciences from U.C. Irvine followed by a Postdoctoral Fellowship at The Scripps Research Institute and U.C. Berkeley. His scientific background includes studies on protein structure-function, natural product biosynthesis, enzymology, oxidation-reduction chemistry, conjugation, and biologics. He currently

leads the protein sciences and process development team at Parvus Therapeutics focused on upstream, downstream, and conjugation processing.

Title: Semi-continuous Processing of Complex Biologics. A Case for Increased Productivity and Quality for Nanomedicine Production
Authors: Chris Warner, Louis Demers, ***Joel Bruegger****, Shu R. Huang, Luis Martinez*

Abstract: Parvus Therapeutics, a Biopharmaceutical Start-up in the Bay Area, is developing Navacims™ for the treatment of autoimmune diseases. These antigen-specific nanomedicines induce immune tolerance through a multi-valent peptide presentation to T-cells after Intra veinous administration. Navacim manufacture involves production and purification of a Peptide Major Histocompatibility Complex (pMHC) through CHO cell culture followed by conjugation to a nanoparticle. pMHC production currently utilizes traditional fed-batch processes yet suffers from low titers. Conjugation reactions can result in low production yields and variable product quality if not precisely controlled. Shifting to continuous processes using perfusion cell culture, real time process analytics to monitor the conjugation reaction, and recycling of unreacted peptide would result in significant advances in cost savings as well as productivity. Critical Quality Attributes must first be identified, defined, and measured. Moreover, control systems must be established to confirm safe and effective product. This talk will present a case study in shifting advanced manufacturing systems from batch processes to continuous with a focus on product quality and regulatory considerations.

Session III – Manufacturing Platforms & Strategies

Session Chair: Chris Hwang, *EVP and CTO, Transcenta*

Irina Ramos, Ph.D.
AstraZeneca

Irina is a Director of downstream continuous manufacturing, in the Bioprocess Engineering and Technology group at AstraZeneca in Gaithersburg, MD, USA. She has been a key driver of the technology development and implementation strategy of continuous clinical manufacturing. Her team works to increase fundamental understanding of the various unit operations of drug substance manufacturing. They work in a cross functional matrix and manage the design, testing and qualification of process equipment and automation on the systems to integrate them. Irina holds a BS/MS in Chemical Engineering from University of Porto (Portugal) and a PhD in Biochemical Engineering from University of Maryland, Baltimore County (UMBC) (USA). She has been involved with the National Institute for Innovation in Manufacturing Biopharmaceuticals (NIIMBL) Process Intensification program by leading the Control Strategy workstream. For the past 13 years, Irina's work in the pharmaceutical industry has spanned from early to late-stage process development and validation, process scalability, CMC and technology transfer to both clinical and commercial manufacturing. Irina has been teaching a class on Downstream Process Development as part of the Professional Master

in Biotechnology from UMBC since 2015. She is passionate about mentorship and developing resources to improve the communication of science.

Title: Evaluation of Fully Automated and Integrated System to Enable Next-Gen Manufacturing for Clinical Trial Material and Commercial Scale

Authors: Nikunj Sharda[1], Zeena Anderson[1], Jared Steffy[2], Darin El-Agib[2], Jon Coffman[1], ***Irina Ramos[1]***

1 Bioprocess Technology and Engineering, Biopharmaceutical Development, AstraZeneca, Gaithersburg, MD, USA
2 Manufacturing Sciences, Biopharmaceutical Development,

Abstract: Next-Generation Manufacturing(NGM) or continuous bioprocessing has the potential to provide significant cost reduction of manufacturing and facility size while improving product quality and providing flexibility for intensified multi-product facilities when compared to the traditional batch mode of operation. The equipment and automation supporting NGM can be complex and expensive. AstraZeneca (AZ)has considered two options: designing its own bespoke NGM system, or buying a pre-built system. PAK BioSolutions provided a turn-key automated and integrated system that can operate four continuous purification stages simultaneously and synchronized. Two PAK system scan be operated in tandem while connected to bioreactor to run the end-to-end downstream process continuously, including dual column chromatography, continuous viral inactivation, in-line conditioning and filtration. The system uses a continuous flowrate to match the upstream perfusion for capture chromatography step, while making sure loading time is greater than regeneration time for the highest titer day to prevent overloading. Additionally, every cycle is loaded to column's maximum capacity (variable loading time), and this provides a robust control strategy for the subsequent unit operations by normalizing the concentrations. Currently a novel virus inactivation is implemented by using an SEC column to achieve the desired constant inactivation time, but a variable in activation time strategy must be considered at commercial scale to achieve significant improvement in vessel size. In this study, the equipment setup was used to purify 200L perfusion bioreactor, running dual column capture, continuous low pH Viral Inactivation, dual column flow-through AEX, continuous in-line CEX adjustment, and dual column bind-and-elute CEX. Throughout the 14-day run, 200 Protein A capture cycles were run processing a total of 3360L of conditioned media with titers ranging from 0.02to 3.0g/L. This resulted in ~4kg of purified CEX product, giving an overall yield>90%. Based on this evaluation, it is very likely that AZ will use this intensified bioprocess strategy for GLP-toxicology material generation as well as early-stage clinical manufacturing.

Christopher Hwang, Ph.D.
Transcenta Therapeutics Inc.

Chris has over 30 years of experience in biopharmaceutical industry, mainly in the areas of CMC development, manufacturing, and support. At Transcenta, he is responsible for developing and implementing advance biomanufacturing platform to dramatically lower cost of goods to expand patient access to innovative biologics. Prior to joining Transcenta in 2016, he spent 25 years at Genzyme and Sanofi, most recently as Senior Director in Late-Stage Process Development and was the program lead for the integrated continuous biomanufacturing (ICB) platform. He also has led a number of clinical and commercial stage products at Genzyme and Sanofi. He received his Ph.D. in biochemical engineering from M.I.T.

Title: Development and Industrialization of Highly Intensified Connected/Continuous Biomanufacturing Platform (HiCB) to Address Patient and Business Needs

Abstract: Industry challenges stem from drug pricing pressure, demand uncertainty, increasing competition, and need for rapid response are driving innovations in biomanufacturing. To achieve the high quality, speed, flexibility, and low cost needed to address these challenges, Transcenta/HJB is developing and implementing novel technologies to achieve our vision of “small and nimble” facility with “output of much larger”.

Here, we present our vision of the future of biomanufacturing which integrates both advanced biomanufacturing platform and facility design, and our 5-year plan (2018 – 2023) to develop and implement this universal production platform in GMP manufacturing. We will share our progress to-date on intensifying our processes and securing the technology needed to support industrialization. From a process intensification perspective, we will share significant progress made in achieving high volumetric productivities and consistent product quality in continuous perfusion processes for multiple cell lines (up to 8 g/L-day) and commercial scale GMP manufacturing. In addition, in close collaboration with MilliporeSigma, highly intensified and automated connected/continuous downstream technologies are being developed and implemented to support the highly productive cell culture platform. We will showcase a single-use integrated capture system and a single-use polishing system “Combo” that connects and automates four-unit operations. These systems are part of the key enabling technologies to debottleneck plant output and are on track for full scale GMP operation in Q1’23. Lastly, we will present our implementation strategy for our pipeline molecules at different stages in development; balancing risks (technical, operational, and regulatory), benefits, and speed of adoption, to ensure speed to clinic and a route to full implementation at commercial scale prior to registration filings to maximize plant output and drive down cost of goods.

Magnus Schroeder
Just - Evotec Biologics, Inc.

Magnus received his M.S. in Molecular Biotechnology with a focus on cell culture from the University Bielefeld, Germany and received his Ph.D. from the Research Center Juelich and University Bielefeld, Germany with a focus on protein purification development. He also conducted post-graduate research at the University of Minnesota and Rensselaer Polytechnic Institute in biochemical engineering and Deakin University, Melbourne, Australia in corporate management.

Title: Advancing Integrated, Continuous Manufacturing in a Flexible cGMP Facility (J.POD®)

Abstract: At Just - Evotec Biologics, we are using an integrated, continuous manufacturing platform to deliver low-cost, high quality biopharmaceutical products as part of our mission to dramatically increase global access to biologics. Our assessment of cost reduction drivers has guided our technology platform towards two key areas: 1) the development of a continuous bioprocessing platform using an intensified, high productivity, longer duration cell culture process, and 2) the design of J.POD®, a flexible and modular cGMP manufacturing facility that leverages single-use technology with a lower cost of capital investment and greater speed of construction. This presentation will discuss the drivers and strategy used by Just - Evotec Biologics to develop an integrated, continuous manufacturing platform which is able to provide both low cost and flexibility for cGMP manufacturing of early/late-clinical phase and commercial biologics. Additionally, the presentation will highlight our first Clinical Ph I through Commercial facility in North America, J.POD 1 US, which launched production in September 2021, and now manufactures antibodies and other therapeutic proteins. Key quality and regulatory considerations associated with process and facility design concepts for operation of a multi-product facility will be highlighted.

Magnus leads Just - Evotec Biologics' Process & Product Design team at the J.POD Redmond, Washington (US) site facilitating FIH, commercialization and technology development programs. Magnus has over 20 years of biopharmaceutical industry experience in process development, CMC strategy, global tech transfer and technology development. Prior to joining Just, Magnus held various roles at Dynavax Technologies, CSL Limited, AGC Biologics and Avid Bioservices. During his career, Magnus has supported the commercial launch of six biopharmaceuticals and numerous IND/IMPD enabling development programs.

Rob O'Keefe
Eli Lilly and Company

Rob Graduated with BEng (Hons) Chemical Engineering Degree from University College Dublin in 2010 and MSc in Biopharmaceutical Science in 2013. He joined Eli Lilly also in 2010 specializing in Bioprocess Design, Facility Design and Start-up, Facility Fit Modelling, and more recently Next Generation Bioprocessing. He has been directly involved in the design, start-up, and validation of Lilly's four mAb facilities and was also the lead process engineer for Lilly's Covid Mab Tech Transfers globally. He is currently finishing a part-time PhD with University College Dublin on Bioprocess Modelling and Next Generation Bioprocessing

Title: Next Generation Bioprocessing (NGB) – Manufacturing and Facility Strategies

Abstract: Over the last decade there has been continuous growth and development in mAb production science and technology. Increases in cell line productivity and improved purification capacity offer a unique opportunity for a major shift in the biopharmaceutical industry towards accelerated product supply, flexibility, high throughput, lower cost per gram, and lower carbon footprint. This research utilises Process Variability Capacity and Operability Modelling to provide several future NGB Concept case studies across a wide range of global mAb supply (1000 – 20000 kg/year). The cases evaluated include:

Base Case: Batch Large Scale Stainless Steel (2 – 5 g/L) – (1000 – 3000 kg/year) – Multi Product (3 Molecules)
NGB_1: Process Intensification Large Scale Stainless Steel (5 – 20 g/L) – (3000 – 20000 kg/year) – (3 Molecules)
NGB_2: Process Intensification Hybrid Single Use Facility (5 – 20 g/L) – (3000 – 20000 kg/year) – (3 Molecules)
NGB_3: Fully Continuous Single Use Facility (1 – 6 g/L/day) – (3000 – 20000 kg/year – (3 Molecules)

This research compares a range of next generation bioprocessing facility concepts, manufacturing strategies, and technologies aiming to clearly demonstrate key benefits, shortfalls, and challenges with each NGB concept. Process Variability Capacity and Operability Modelling is utilized to assess the throughput capability, flexibility, operability, facility logistics, environmental sustainability, and economic viability of each concept across a range of mAb supply requirements (kg/year), highlighting the impact of annual demand.

Day 2 - Thursday, December 8, 2022

Session IV – Regulatory Validation and Considerations

Session Chair: Kristina Pleitt, Sr Manager, Bioproduction R&D Innovation, Thermo Fisher Scientific

Ioana Pintescu
Asahi Kasei Bioprocess America

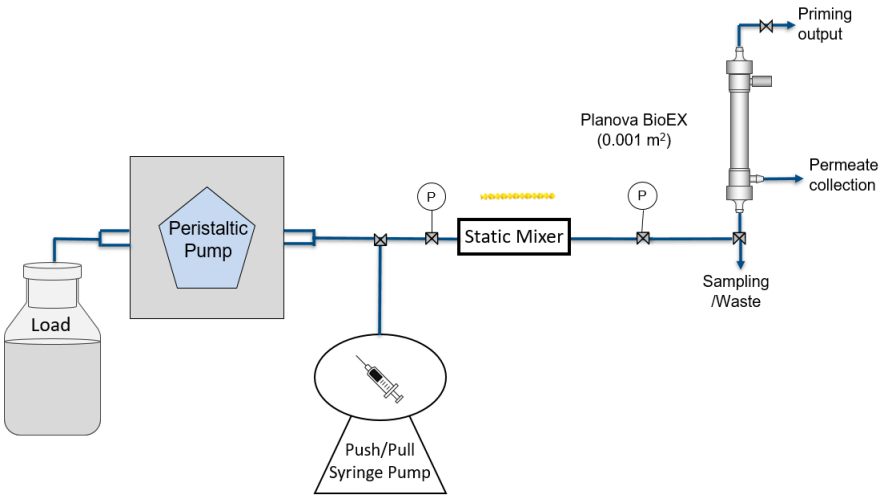
Ioana is a Research Associate in the Research & Development group at Asahi Kasei Bioprocess America located in Glenview, IL. She joined the company in 2020 after completing her B.S. in Molecular and Cellular Biology from the University of Illinois at Urbana-Champaign. While at school, she was an undergraduate research student in the Harley Lab, a chemical and bioengineering lab where she spent two years developing a three-dimensional model of the menstrual cycle. In her current role, Ioana's responsibilities include performing viral clearance studies as well as overseeing lab safety.

Title: Designing a Small-Scale Model to Test Continuous In-Line Spiking in Virus Filtration

Abstract: Virus filtration today is primarily performed by traditional batch mode processing and is typically performed under constant pressure and for a limited duration. There have been many efforts to shift downstream product purification steps to continuous bioprocessing as these technologies have the potential to increase productivity and product quality.

For viral clearance validation of traditional batch processes, load solution is spiked with virus and filtered through the virus removal filter. The goal of this study is to develop a small-scale model for in-line spiking in continuous virus filtration. While there are many differences between batch mode and continuous mode processing, this project is focused on establishing an in-line virus spiking method over a long-duration (high throughput) filtration under constant flow mode. Understanding these differences can help improve validation strategies for future large-scale processes.

Bovine serum albumin (BSA) solution, which was continuously spiked with PP7 bacteriophage, was loaded onto a 0.001 m^2 Planova™ BioEX filter. A push/pull syringe pump was used to continuously spike virus into the system. This was then followed by mixing the spiked load solution using a static mixer prior to reaching the virus filter, as shown in the schematic of the set-up below.



Preliminary work included testing the validity of in-line solution mixing. Conductivity of salt solutions and absorbance of BSA solution showed sufficient in-line mixing. Subsequent testing included determining conditions that can be used for a continuously spiked filtration. A 24 h PP7-spiked filtration run showed complete viral clearance in the filtrate sample and successful PP7 and BSA mixing by the in-line mixer. Future studies will focus on longer filtration durations and introducing mammalian viruses and other representative molecules such as monoclonal antibodies. The findings from this project will aid in understanding and overcoming the technical challenges associated with implementing continuous virus filtration in biomanufacturing.

Joel Welch
FDA

Joel Welch is the Associate Director for Science& Biosimilar Strategy in the Office of Biotechnology Products in the Office of Pharmaceutical Quality at the US Food and Drug Administration. He is responsible for assessing emerging, complex, or precedent-setting issues impacting science policies of the office with particular emphasis on the biosimilar program. He also serves as the Rapporteur for the ICH revision to Q5A(R1) and the Chair for the Emerging Technology Program. In his time at FDA, he has also served as a Review Chief, Team Leader, Primary Assessor and Regulatory Project Manager. Prior to joining FDA, he spent 6 years in industry supporting late state analytical development of small molecules.

Title: Opportunities in Continuous Manufacturing and the Emerging Technology Program

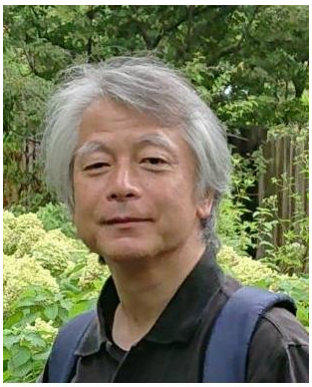
Abstract: This talk will provide a discussion on the benefits of Advanced Manufacturing (including Continuous Manufacturing) and the US-FDA perspective. This perspective will also include an overview of the Emerging Technology Program, its collaborative approach, the program objectives, and the lifecycle of a novel technology. The discussion of the lifecycle will include a brief overview of the approach to technology graduation. Finally, suggestions on how to engage with the ETP and recent trends in submissions and novel technology will be provided

Scott Lute
US FDA

Scott Lute is a Senior Biologist and Product Quality Assessor in the Office of Biotechnology Products, Division 2 and a member of the CDER Manufacturing Science & Innovation Center of Excellence. He has been with CDER/OBP since 2002. Over the past 20 years he has extensively studied various aspects of monoclonal antibody manufacturing focusing on regulatory concerns associated with the viral safety of downstream bioprocessing. His research has supported the development of PDA Technical Reports and the establishment of recent "Recommended Practices" for OBP, providing a knowledge base for FDA assessors. His current research focus includes advanced manufacturing technologies with a focus on continuous manufacturing to provide regulatory support for the Emerging Technology Program submissions at CDER.

Title: Viral Clearance Strategies for Continuous Manufacturing

Abstract: Continuous manufacturing of biologics has rapidly evolved over the past several years. The implementation of novel equipment and novel strategies to replace batch operations requires research to understand the impact of such changes on the viral safety of the continuous process. This talk will focus on the continuous viral clearance strategies for three key unit operations: continuous capture, continuous viral inactivation, and continuous viral filtration. Summaries of published CDER, industry, and vendor research will be discussed to demonstrate a general understanding of critical aspects/risks that may be associated with these key unit operations. By understanding the risks, one can design representative small scale models for viral clearance studies, ensuring the viral safety of the continuous manufacturing process.



Hironobu Shirataki, Ph.D.
Asahi Kasei Medical, Co., Ltd.

Dr. Shirataki graduated physics department of Hokkaido University in Japan and got the Doctor of Science by the study of polymer physics. He has a lot of carriers in the research and development as an employee of Asahi Kasei Co., ltd. Currently he is a Senior consultant of scientific affairs at Asahi Kasei Medical and mainly studying the virus filter and column chromatography for the protein purification and virus reduction.

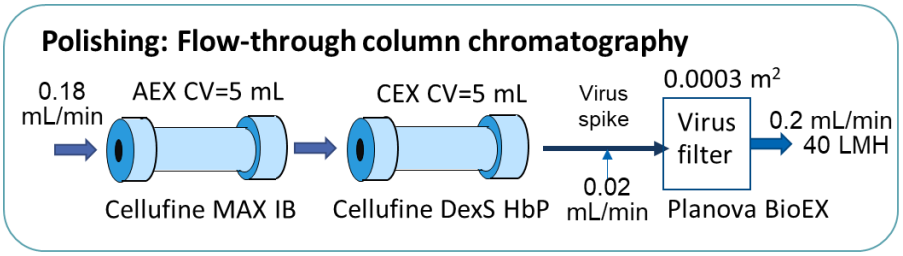
Title: Constant Flow Rate Viral Clearance Study of Planova™ BioEX Virus Removal Filter and Implementation into an End-to-End continuous Process for mAb Purification

Abstract: Virus filtration is located at the end of downstream processing and is a critical step in removing viruses from biopharmaceutical products. However, research and technological developments for integrating virus filtration into continuous processes are still ongoing. One suggested solution is integrating filters after column chromatography processes, but there are challenges such as balancing throughput capacity and flow rate across the system, process pauses affecting virus filter robustness and pressure stability in long duration filtrations. We investigated the filtration behavior and virus removal capability for a monoclonal antibodies (mAb) process in an integrated column chromatography and virus filtration setup under constant flow rate.

Mixed-mode AEX and modified CEX columns were run in-series in a pool-less integrated setup with a 0.0003 m^2 Planova BioEX virus filter connected, as shown in the figure (upper left). In order to investigate filter robustness, viral clearance tests for the filter were conducted using the inline spike test method. To address the concern of virus breakthrough, another test with a 60-min process pause included between mAb solution load and recovery flush was conducted during the virus filtration step.

A solution of 10 mg/mL mAb with quantified host cell protein (HCP) content that was processed in the integrated setup achieved high HCP reduction (2.94 ng/mg-mAb) and high mAb recovery (94%). Inline virus spike test using the same solution at 40 LMH (0.2 mL/min) constant flux had robust MMV virus reduction as shown in the table below. The two column in-series process also showed high virus reduction, with tests for MMV yielding a virus LRV greater than 5 even at a flow rate less than 5 LMH for Planova BioEX filters.

Thus confirmed integrated setup connecting 2 columns and virus filter directly was implemented into the End-to-End continuous process as shown in the figure (down). In the process, batch virus inactivation was executed with auto pH control to clarify the production batch concept. The stable mAb continuous process from perfusion cell culture to virus filtration with high purification and recovery was demonstrated by this process.



	MMV
Starting material [log_{10}]	7.54
LRV of flow through fraction	≥ 5.47 ± 0.25
LRV of flow through fraction + wash after 60 min process pause	≥ 5.07 ± 0.24

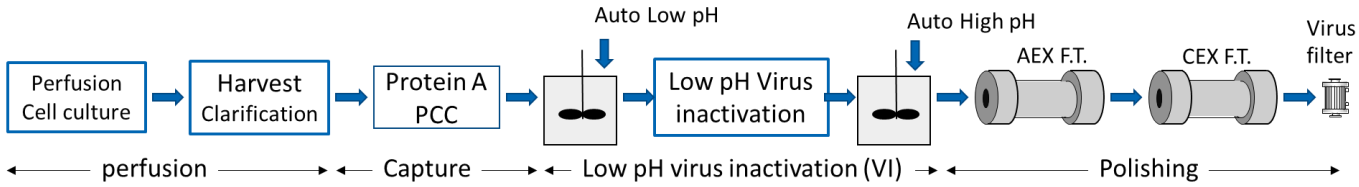
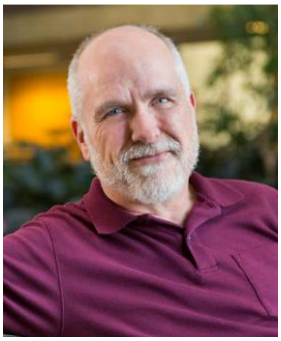


Figure 1. Schematic of flow-through two-column chromatography and virus filter integrated system (upper left), virus removal results across the filter (upper right) and End-to-End continuous process setup (down).

Session V – Technologies to Advance Continuous Manufacturing

Session Chair: Rich Chen, Exec Dir Purification & Viral Safety, Eli Lilly and Company



John Moomaw
Eli Lilly and Company

John Moomaw is currently a Senior Principal Chemist at Eli Lilly & Company. He earned his Bachelor of Science in Biology at SUNY Stonybrook, NY in 1983 and began work at Cold Spring Harbor labs. John received his MS in Genetics from the University of North Carolina at Chapel Hill in 1987. After a stint as an auto mechanic and shop manager he joined the lab of Patrick (Pat) Casey at the Duke University Medical Center serving as a technician and lab manager from 1990-1998.

John joined Lilly from Duke University in 1998 to work in discovery research (Infectious Diseases). He transitioned to the purification process development group in 2002 to support Lilly's emerging monoclonal antibody therapy (mAb) platform. From there John led and enabled several mAb projects and tech transfers to commercial sites. He began a deep focus on UF/DF in 2015 and has become the TFF subject matter expert.

Title: Continuous TFF Development: Equipment, Process, and Process Control

Abstract: Multiple publications and ongoing efforts are focused on the implementation of continuous tangential flow filtration (cTFF) technologies within the biopharmaceutical industry. Single pass TFF (spTFF) for concentration, and counter-current buffer exchange TFF (CCBE-TFF) for diafiltration, are examples of these technologies. Potential benefits of cTFF relative to batch processes include reduced pump passes for shear-sensitive products, reduced buffer consumption, and incorporation into continuous downstream processes. Potential challenges include complex equipment layouts, complex process control strategies, more equipment/instrumentation, and process optimization relative to batch systems.

This work is part of an on-going program at Lilly to progress the conceptual design of a next generation bioprocessing manufacturing facility for high throughput assets. Continuous technology is well-suited to achieving high throughput with a limited footprint, however, continuous solutions for commercial TFF unit operations in the pharmaceutical industry are limited. Here, we share Lilly's development efforts to design a fully integrated/automated lab/pilot scale cTFF system for continuous concentration and buffer exchange of monoclonal antibodies (mAbs) or other solutes of interest.

Lilly's cTFF design is the result of a partnership between Development and Engineering groups built upon foundational experimentation, process and process control engineering first principles, and proof-of-concept experimentation. The design utilizes a series of integrated spTFF modules to perform both concentration and counter-current buffer exchange for a given molecule/process in a continuous flow path. A process control strategy is defined to achieve robust steady-state operation across the multistage integrated cTFF system.

A standardized approach for filter device selection and operating parameter optimization is established based upon the targeted use of spTFF flux-excursion studies. The design has been demonstrated at laboratory scale achieving greater than 99.9% buffer exchange and mAb concentrations up to 250 g/L. Steady-state operations as defined by stable flow, pressure, conductivity, and concentration have been demonstrated for multiple hours of run time.

Daniel Some, Ph.D.
Wyatt Technology Corp. (on behalf of BioPhorum, Technology Roadmapping Phorum, ILM/RTR Workstream)

Dr. Daniel Some is Principal Scientist at Wyatt Technology Corp., where he contributes to product and application development as well as scientific and technical marketing. Currently he leads Wyatt's program for commercializing real-time MALS PAT products. Dan studied undergraduate physics at the Technion - Israel Institute of Technology and obtained his Ph.D. in physics from Brown University, then carried out postdoctoral research at Los Alamos National Laboratory and the Weizmann Institute of Science. Prior to joining Wyatt he was involved in defense technology and the development of processed wafer inspection tools for the semiconductor industry.

Title: Towards Automated Pooling Based upon Real-time Aggregate Detection during Cation Exchange Bind-and-Elute Chromatography: Phase 1 Proof-of-Concept

Abstract: Real-time process analytics are essential for effective and robust downstream processing operations in the continuous manufacturing (CM) of monoclonal antibody-based therapeutics (mAbs). To date, real-time quantification of mAb aggregates, needed for mAb polishing steps, has proven challenging, with traditional process analytical technology (PAT) instrumentation such as UV/Vis spectroscopy or Raman scattering insufficiently sensitive. BioPhorum's Technology Roadmapping In-line Monitoring/Real-time Release workstream has identified real-time aggregate monitoring as a key PAT objective with significant potential for increasing mAb yield and improving product quality. The workstream has selected real-time multi-angle light scattering (RT-MALS) as the most promising technology for meeting these goals.

Flow-through chromatography is a common polishing process in CM. The utility of RT-MALS for monitoring mAb aggregates and controlling pooling in flow-through hydrophobic interaction chromatography has been demonstrated by Patel et al. (*mAbs*, 2018). In traditional batch processing of mAbs, bind-and-elute cation exchange (B/E CEX) chromatography is the most common method for aggregate removal, while column-switching B/E CEX is an alternative to flow-through chromatographic polishing in CM. Standard approaches to B/E CEX pooling require relatively large safety margins in order to maintain acceptable final aggregate levels when input material quality or process conditions vary. It is expected that real-time aggregate quantification will enable tighter safety margins with no detrimental impact on aggregate content in the final pool.

This talk will discuss in-line / on-line RT-MALS, its potential for monitoring mAb aggregates, the challenges RT-MALS faces in a bind-and-elute CEX process, and how these have been tested and demonstrated in Phase 1 of a proof-of-concept study carried out under the auspices of BioPhorum. An aspirational goal of this proof of concept is improving monomeric mAb yield by at least 5% while maintaining product quality.

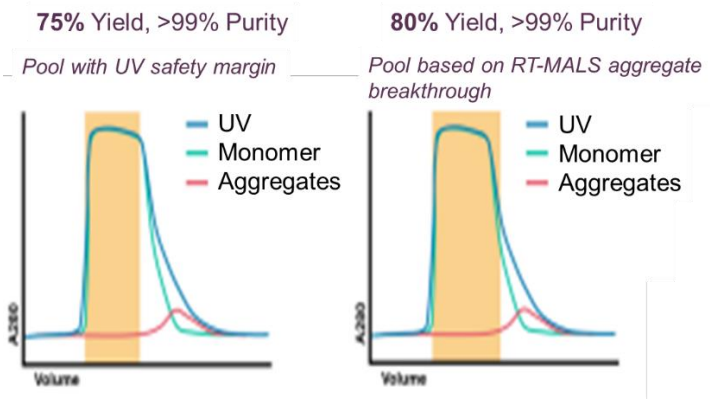


Figure 2. Illustration of aggregate monitoring proof-of-concept goal

Jens Poschet
MilliporeSigma

Jens Poschet is Portfolio Project Manager Department MAST and has been working in biosciences for over thirty years in various positions at academic institutes, government and industry. He is currently employed by MilliporeSigma and is part of the Modular Automated Sampling Technology team. He will present data generated by MilliporeSigma in Burlington, MA.

Title: Utilization of Automated Aseptic Sampling for Accelerated Access to Process and Quality Data in Upstream Bioprocessing

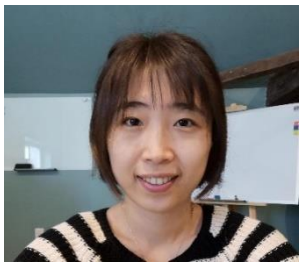
Abstract: Interest to incorporate more Process Analytical Technologies (PAT) in bioprocess increased in the recent years. This development is important to enable increased process understanding and transforming the way we look at the process and how we analyze the data.

At MilliporeSigma (A business of Merck KGaA, Darmstadt, Germany) we target the following PAT initiatives:

- Aim to monitor all relevant process indicators and CQAs in a real-time or timely manner,
- Aim to retro-control the process in a continuous manner
- Aim to accelerate batch release

In this context, the team is identifying, evaluating, and deploying the right technologies in both process development with the final aim to deploy these methodologies in clinical manufacturing. As of today, we cover the different process indicators and CQAs. Currently, the team is working on a combination of on-line and in-line sensors, automated sampling and at-line methods, with the final aim to have these events being orchestrated

using a PAT-manager software component. Here, we will be focusing on presenting/evaluating results that compare manual sampling process versus automated sampling using the Modular Automated Sampling Technology (MAST®). We will also present further data obtained that supports the strategic aspects and challenges of this transformation journey.



Rui Wheaton, Ph.D.
National Resilience, Inc.

Rui Wheaton is a modeling & simulation scientist, and currently leading process economics and process system modeling program at Resilience.

Rui has a wealth of experience in process engineering, computational fluid dynamics (CFD), and mechanistic modeling, and have used these tools for process performance and comparability evaluation, root cause investigation, and process improvement.

Rui received her PhD in Chemical Engineering from Worcester Polytechnic Institute. Prior to joining Resilience, she has worked in Process Engineering roles at Bristol Myers Squibb and Praxair and has applied her expertise to support process development, scale-up and troubleshooting of commercial processes.

Ahsan Munir
National Resilience, Inc.

Ahsan Munir is a Director of Process Modeling & Data Science at Resilience. He is leading the development of engineering digital solutions, by integrating computational fluid dynamics (CFD), mechanistic and data-driven modeling approaches for monitoring, optimization, control and scale-up of bioprocesses.

A thought-leader in digital innovation, Ahsan has extensive experience in modeling and simulation in a variety of biopharmaceutical spaces including cell & gene therapy, vaccines, medical devices, and protein therapeutics. His skill sets also extends to a broader array of chemical engineering process modeling and design.

Ahsan received his PhD in Chemical Engineering from Worcester Polytechnic Institute and has honed and advanced his skills at COMSOL, Amgen, and Vertex Pharmaceuticals. He has successfully applied his expertise in support of process

development, scale-up, risk assessment, troubleshooting of commercial processes, and in next-gen technology evaluation.

Title: A Digital Twin of an Integrated and Continuous Biomanufacturing (ICB) Process at Resilience

Authors: Rui Wheaton, Ahsan Munir, Chris VanLang, Thomas Erdenberger, Huanchun Cui, Brian To, Thomas Ransohoff

Abstract: National Resilience, Inc. (Resilience) is a first-of-its-kind manufacturing and technology company dedicated to broadening access to complex medicines and protecting biopharmaceutical supply chains against disruption. As a part of this vision, Resilience is developing an advanced end-to- end continuous drug substance manufacturing platform for biological molecules. To enable robustness, facilitate *in silico* process characterization, and enable an efficient process control strategy, we are developing a process digital twin of an integrated and continuous biomanufacturing (ICB) process at Resilience. The ICB digital twin uses Resilience advanced computational modeling framework together with a data-driven and mechanistic modeling approaches. Two objectives of the digital twin model are to add significant value across the process lifecycle by ensuring effective management of process deviations and by reducing the amount of experimentation to expand process knowledge and optimize operating conditions.

Described herein are the steps for the development of ICB digital twin model, the individual unit operation process models were built separately and then integrated into an end-to-end flowsheet simulation. The model is being used to determine the impact of expected disturbances, deviations, and uncertainties on product quality. The goal is to use residence time distribution analysis to identify the duration of product diversion in response to the deviation, and allow product impacted by disturbance to be diverted without impacting the reminder of the batch.

The ICB Digital Twin will enable us to characterize and build a robust process control strategy with fewer physical experiments, reduced risk, and fewer pilot trials. It will help us to successfully predict process performance and product quality in real time and enable us to do the virtual exploration of a process design space facilitating data-driven science- and risk-based implementation of integrated continuous biomanufacturing (ICB) process at Resilience.