Final Agenda

Register by June 4 and SAVE UP to $350!

THE BIOPROCESSING SUMMIT

Cambridge Healthtech Institute's Second Annual

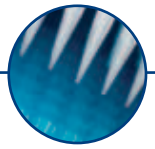
Practical Solutions for Today's Laboratory Challenges

August 23-26, 2010

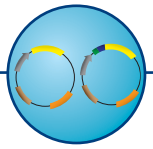
Marriott Long Wharf Hotel, Boston, MA

AUGUST 23-24

Optimizing Cell Culture Technology

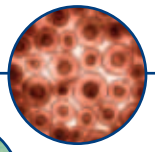


Affinity Tag Protein Purification



AUGUST 25-26

Optimizing Mammalian Cell Lines



Baculovirus Technology

Short Courses:

AUGUST 23

Optimizing Media – *Achieving Super Soup*

Affinity Tag Purification Systems

AUGUST 24

Dinner Short Course: *E.coli* Innovations

Corporate Sponsors

BlueSky Biotech · cevec · forteBIO · IBA BioTAGnology · PALL Life Sciences *New Science. New Thinking.SM* · Promega

BioprocessingSummit.com

Organized by CHI Cambridge Healthtech Institute
250 First Avenue, Suite 300 • Needham, Massachusetts 02494 • Telephone: 781-972-5400 • toll-free in the U.S. 888-999-6288 • Fax: 781-972-5425

Hotel & Travel Information

CONFERENCE VENUE AND HOTEL:

Marriott Long Wharf Hotel
296 State Street, Boston, MA 02109
T: 617-227-0800 • F: 617-227-2867

Discounted Room Rate: $179 s/d
Discounted Room Rate Cut-off Date: July 26, 2010

Please call the hotel directly to reserve your sleeping accommodations. Identify yourself as a Cambridge Healthtech Institute conference attendee to receive the reduced room rate. Reservations made after the cut-off date or after the group room block has been filled (whichever comes first) will be accepted on a space-and-rate-availability basis. Rooms are limited, so please book early.

FLIGHT DISCOUNTS:

To receive a 5% or greater discount on all American Airline flights please use one of the following methods:

- **Call 1-800-433-1790 (authorization code 4680AA).**
- **Go online at www.aa.com (enter 4680AA in promotion discount box).**
- **Contact our designated travel agent, Wendy Levine, at 1-800-336-5248 ext. 137.**

CAR RENTAL DISCOUNTS:

Special discount rentals have been established with Hertz for this conference. Please use one of the following methods:
Call HERTZ directly at 800-654-3131 OR go online www.hertz.com use our Hertz Convention Number (CV): 04KL0001

Sponsorship & Exhibit

AGENDA PRESENTATIONS

Includes a 15 or 30-minute presentation during a conference program of your choice, ensuring that your audience is seated and ready to hear your talk.

BREAKFAST OR LUNCHEON PRESENTATIONS

Includes a 30-minute presentation given over breakfast or lunch, which is served on behalf of your company. Workshop is concluded with a 15-minute Q&A session, allowing further interaction with your customer base. CHI can customize a sponsorship to meet your needs and budget. We offer comprehensive packages that give your company exposure before, during and after the event. Sponsorships include a talk, exhibit space, conference registrations, wide-ranging branding opportunities, use of event mailing lists and more.

ADDITIONAL SPONSORSHIP & PROMOTIONAL OPPORTUNITIES

- Invitation-Only VIP Functions
- Chair drops of company literature (program specific)
- Branded badge lanyards
- Branded conference tote bags
- Branded attendee coffee mugs
- Branded padfolios

EXHIBIT HALL

This event will host more than 300 of your hard to reach end-users. Promote your latest technology, solution or service directly to qualified buyers!

FOR MORE INFORMATION PLEASE CONTACT:

Suzanne Carroll
Manager, Business Development
781-972-5452
scarroll@healthtech.com

Media Partners

Lead Sponsoring Publications

Pharmaceutical Review
Bio-IT World
BioPharm International
GEN Genetic Engineering & Biotechnology News
International DrugDiscovery
nature
Science AAAS
The Scientist

Sponsoring Publications

ddn Drug Discovery & Development
PharmaVOICE

Web Partners

BioSpace.com
www.PharmCast.com – Pharmaceutical Internet

Short Courses*

Pre-Conference Short Courses
Monday, August 23 • 9:00am to 12:00pm

(SC1) Optimizing Media – *Achieving Super Soup*

To grow mammalian cells, researchers need to provide an optimal *in vitro* environment. The key feature of successful cell growth is the culture medium. 'Achieving Super Soup' requires finesse and know-how in order to combine the right ingredients at the right times under the right conditions to achieve high titers. This workshop will provide a foundation for optimizing cell culture media presented by real-world experts who will also tailor a portion of the course to fit concerns and challenges faced by the workshop participants.

Short Course Topics:

- Details of nutrient limiting technique
- Large scale application of technology for clinical product
- Minor upstream impacts of technology

Monitoring Metabolic Parameters in Cell Culture Processes

Timothy Fawcett, Ph.D., Director, The BioTechnical Institute of Maryland, Inc.

Cell culturing conditions play an important role in providing a suitable environment for the growth and development of animal cells. As expectations for productivity and reproducibility increase, a closer examination of mammalian cell culture media components is necessary. Therefore, we will discuss key media components, their reason for inclusion in media and the positive and negative results of decay, and metabolic breakdown. A rationale for monitoring some key media constituents and metabolites will also be given along with suggestions to enhance culturing conditions and increase productivity.

Customizing Cell Culture Media and Reagents

Michael Hanson, Director, Media Division,Caisson Labs

Sourcing customized cell culture media and reagents can provide unique challenges for individual consumers. This talk will explore the feasibility of producing custom media as well as the cost and other considerations faced by end users. Case studies will be presented to demonstrate some of challenges of custom manufacturing and ways those challenges have been overcome.

(SC2) Affinity Tag Purification Systems

Affinity tagging of recombinant proteins allows the purification of multiple proteins with one technique. In order to provide efficiency it is important to explore the limitations and opportunities, various generations of purification systems have to offer. This workshop provides the opportunity to learn about manual and automated purification systems, to assess the challenges and advantages involved and to discuss common problems during purification.

Short Course Topics:

- Overview over current systems
- Advantages and challenges of systems
- Multistep purifications
- Automated systems – an advantage?
- Cleavage of affinity tags
- Addressing common problems during protein purification

Dinner Short Course
Tuesday, August 24 • 6:15pm to 9:00pm

(SC3) *E.coli* Innovations

Escherichia coli has proven its worth as a protein expression platform. Currently, *E.coli* is not viewed so much as an 'alternative' platform, but as a viable choice for achieving high-level expression of human genes and protein. This Dinner Short Course will explore strategies for successful *E.coli* protein expression, including:

- Host cell engineering to improve product quality,
- Development of protein expression assays,
- Development and optimization of protein purification processes,
- Automating high-throughput protein expression and purification, and
- Scaling-up production

Optimizing Expression in Escherichia coli: Conditions and Strains and Hosts, Oh My

Dominic Esposito, Ph.D., Principal Scientist, Protein Expression Lab, SAIC Frederick

Expression of proteins in E. coli requires optimization of a large number of variables in order to obtain the highest levels of good quality protein. As targets become more challenging, considerably more effort is required to find optimal conditions for expression. This talk will discuss many of the variables that can be optimized for improved expression, and ways to minimize the time and effort required to find the best conditions for a given protein. We will examine the effect of different host strains, conditions for expression, and the use of expression and solubility fusion tags on the production of large amounts of useful protein from E. coli.

- E. coli expression technologies
- strain selection
- IPTG vs autoinduction
- solubility enhancement using tags
- ways to improve optimization throughput

E.coli Expression Optimization Methodologies

Rebecca Page, Ph.D., Assistant Professor and Principal Investigator, Department of Molecular Biology, Cell Biology and Biochemistry, Brown University

**Separate registration required*

Optimizing Cell Culture Technology

Enhancing the Environment for Growing Cells

August 23-24, 2010

MONDAY, AUGUST 23 - DAY ONE

12:00pm Conference Registration

OPTIMIZING CHO

1:30 Chairperson's Remarks

Susan Windham-Bannister, Ph.D., President & CEO, Massachusetts Life Sciences Center

1:40 Opening Keynote Presentation:

Strategies for Evolving a CHO-Based Expression Platform

John E. Mott, Ph.D., Director, Bioprocess R&D, Cell Line Development & Global Biologics, Pfizer, Inc.

There are a multitude of diverse challenges facing scientists working in the mammalian-based biotherapeutic protein expression field today. Expectations for shortening development timelines, decreasing resources, increasing volumetric titers and reducing the cost of clinical manufacturing continue to intensify. This presentation will review the various strategies used by Pfizer's Cell Line Development group in order to meet these expectations. The presentation will describe a CHO-based monoclonal antibody platform that uses ACF media, automation, cell line engineering and process technologies that enable the rapid identification of cell lines to be used in clinical and commercial manufacturing.

2:10 Improving Biotherapeutics Through the Use of RNAi

David Kocisko, Ph.D., Principal Scientist, Alnylam Pharmaceuticals

RNAi technology has the ability to improve biotherapeutic manufacturing by selectively and potently turning off problematic genes in biologic producing cell lines. siRNAs target a specific mRNA sequence in the gene of interest to promote its degradation and reduction of corresponding protein synthesis. One of the many strengths of this approach is that siRNAs can be used with any existing cell line, including previously banked cell lines, as an additive to the culture media without any need for cell engineering. In addition, multiple genes can be simultaneously knocked down using a mixture of individual siRNAs directed against each gene target. Using DG44 CHO cells as a model system, siRNAs added directly to the media reduced Bax, Bak, and LDH protein levels by >90% and nearly doubled the integral cell area. Efficient uptake of siRNAs in 40L bioreactors containing DG44 CHO cell cultures has been demonstrated. Finally, siRNA approaches to manipulate other important pathways are currently being developed and demonstrated in DG44 cells.

2:40 Development of a Glutamine Synthetase (GS) Knockout CHO Host Using Zinc Finger Nucleases

Lianchun Fan, Ph.D., Research Scientist, BioProcess Research & Development, Eli Lilly and Company

Over the past decade, the GS-CHO expression technology has been applied successfully for the production of therapeutic proteins, and in particular, monoclonal antibodies. Selection of recombinant cell lines is based on expression of the gene coding for the glutamine synthetase (GS) enzyme, which is introduced by the expression plasmid. However, parental host cells possess an endogenous copy of the glutamine synthetase gene, which can impact the selection process. In order to eliminate the endogenous GS basal expression and attempt to tighten selection of the transfected population of cells, we utilized zinc finger nucleases targeted against the GS gene to disrupt the genomic loci. The resulting cell lines were characterized to confirm this gene disruption and subsequently evaluated for their performance in the cell line generation process. Based on these studies, the GS knockout cell lines resulted in the elimination of non-producing cells and a significant shift in the mean productivity of the transfected cell population, thus providing for a much more efficient cell line generation screening process.

3:10 Sponsored Presentation *(Opportunity Available – Please contact Suzanne Carroll at scarroll@healthtech.com)*

3:30 Networking Refreshment Break with Exhibit and Poster Viewing

4:15 Moderated Small-Group Breakout Discussions

Join with your colleagues to discuss the crucial issues associated with culturing mammalian cells and purifying proteins using affinity tags. Small-group discussions are a great way to network, exchange information, and develop collaborations. Each table topic is facilitated by an expert in the field, and discussions can be lively and spirited.

1. Making a Healthier Cell Line

Moderator: Lianchun Fan, Ph.D., Research Scientist, BioProcess Research & Development, Eli Lilly and Company

This discussion will mainly focus on the host cell engineering work that deals with anti-apoptosis CHO cell line development.

- Current strategies to develop anti-apoptosis cells
- Best targets to work with
- Feasibility for new cell lines to be implemented into manufacture process

2. Alternative Expression Systems

Moderator: John H. Chon, Ph.D., Senior Director, BioProcess R&D, PERCIVIA, LLC

Chinese hamster ovary (CHO) cells are undisputedly the most widely used of all cell lines in industry for the expression of recombinant proteins for human therapeutics today. Being a very conservative industry, the biopharmaceutical world has thus far been reluctant to explore and utilize other cell lines despite some obvious advantages over CHO cells. However, as we are now well into the 21st century, more and more alternative cell lines that address some of the shortcomings of CHO cells are now gaining wider acceptance. This breakout session will discuss these alternative expression systems and what they bring to the table with respect to safety, quality, and productivity.

- What are the pros and cons of using CHO cells for human therapeutics manufacturing? Are any of the cons show stoppers?
- What alternative cell lines are available to address the "cons" of CHO cells?
- What are the (perceived and proven) risks of using non-CHO cell lines? What would be needed to overcome these risks?
- What is the "dream cell line"?

3. From Bench to Bedside: Cell Culture's Journey from the Lab to the Clinic

Moderator: Sajjad Ahmad, Ph.D., Academic Clinical Lecturer, Institute of Human Genetics and North East England Stem Cell Institute, Newcastle University

Cell therapy, for example involving stem cells, is becoming increasingly discussed and indeed used as a means for curing diseases. Bone marrow transplantation is the classic historic example. The aims of this workshop are to discuss how cells processed or cultured in the laboratory can be taken forward to the clinic for therapeutic applications. The following areas will be discussed:

- Good manufacturing practice
- Regulatory challenges
- Scaling up of cells
- Transportation of cells

4. Improved Early Screening of Cell Lines

Moderator: Brian Majors, Ph.D., Scientist, Clinical Cellular Engineering, Biogen Idec

Cell line development groups have made great strides in production cell line titers. A challenge now is to find methods of early prediction of cell line production and product quality characteristics. This small-group discussion will examine high-throughput screening technologies in cell line development, including:

- automation,
- deep well culture, and
- high-throughput analysis.

5. Cell Bank Stability: What Does It Mean and How Do You Measure It?

Moderator: Jim Moldenhauer, M.S., Senior Research Scientist, Central Cell Banking, Manufacturing Science & Technology, Eli Lilly & Company

The ICH Topic Q 5D, "Quality of Biotechnological Products: Derivation and Characterisation of Cell Substrates Used for Production of Biotechnological/Biological Products", provides guidance on demonstrating stability of cell substrates both during production and storage. This guidance is found in section 2.3.3, "Cell Substrate Stability", and is concerned with evaluation of cell line (or strain) stability during cultivation for production as well as

providing evidence for stability of cell banks during storage. As cell banks have come under increasing regulatory scrutiny, this guidance is referenced during every regulatory agency inspection, and is part of every regulatory submission for product marketing. Therefore, compliance with this guidance is paramount to commercial manufacturing of biological products. However, there is no roadmap to success and approaches will vary on how to meet the spirit of these guidelines. This break-out session will initiate a dialogue between participants on how they believe 'cell substrate stability' should or could best be demonstrated in the context of ICH guidance.

6. Cell Culture Supplements: Quality Aspects

Moderator: Fouad Atouf, Ph.D., Scientific Liaison, The United States Pharmacopeial Convention

One of the challenges associated with the development of therapeutic products derived from cell culture is the use of reagents and ancillary materials as cell culture supplements. These types of reagents can be of animal origin, are not intended to be in the final products and their removal from the finished product is critical for the quality of this category of therapeutics. This small-group discussion will focus on:

- Qualification of cell culture supplements
- Animal-derived vs. animal-free materials
- Residual testing

7. Dealing with Cell Death in Mammalian Cell Cultures

Christopher D. Gregory, Ph.D., CSO, ImmunoSolv Limited; Professor, Inflammatory Cell Biology; Deputy Director, MRC Centre for Inflammation Research, Queen's Medical Research Institute, University of Edinburgh

Cells die for a variety of reasons in culture, from accidental to deliberate (following drug selection, for example). Dying and dead cells affect their culture microenvironment in different ways and cell death can be a significant limiting factor in effective production, storage and delivery of cells for multiple purposes including bioprocessing, R&D and therapy. This breakout session will discuss issues surrounding death of cells in vitro and measures to minimise its detrimental effects.

- Causes of cell death in culture
- Assessing cell death
- Phases of cell death and impact on the cell culture microenvironment
- Dead cell removal from cultures
- Cell death during cell storage and transport

5:15 Roundtable Report Out – Each topic leader will present a brief report of the group's discussion to the meeting delegates.

5:30 Reception in the Exhibit Hall *(Sponsorship Available)*

6:30 End of Day One

TUESDAY, AUGUST 24 - DAY TWO

7:30am Morning Coffee *(Sponsor Opportunities Available)*

OPTIMIZING THE PROCESS

8:25 Chairperson's Remarks

8:30 Development of Robust and Predictive Fed-Batch Scale-Down Models for Mammalian Cells Producing Recombinant Proteins

Arnaud Perilleux, Scientist, Cell Culture, Upstream Process Development, Merck Serono SA

This presentation focuses on the development of predictive and robust fed-batch cell culture scale-down models for several recombinant cell lines. The process performs reproducibly and scales faithfully from the 96 multi-well plates to the 5KL bioreactor scale. A variety of techniques to optimise process parameters, conditions, and medium composition at the different scales are presented, including high-throughput cell culture, metabolic pathway studies and computational fluid dynamic (CFD) technology. Optimized fed-batch process for several recombinant cell lines exhibited significant lactate consumption when the cells entered the stationary or death phase, and enhanced product glycosylation. The ability to automate culture operations and to run multiple wells in parallel that accurately represent large-scale bioreactor process performance in terms of metabolic profiles and product quality, offers considerable enhancement in experimental throughput and flexibility.

9:00 Practical Applications of Design of Experiment (DoE) in Cell Culture

Brigitte Van der Haegen, M.D., Ph.D., Partner & Consultant, Cell Culture Solutions LLC

This presentation will demonstrate the advantages of using DoE in the development of cell culture processes. We will illustrate how to select vital factors using designed screening procedures, how critical factors interact and affect a process and how to determine the right factor settings for optimum performance. We will also address the use of DoE in formulation design. Selected designs of interest to cell culture will be discussed during the presentation.

9:30 Use of Twenty-Four Well Micro Bioreactor as a Scale-Down Model for Optimizing Cell Culture Process Development

Mark Kwatia, M.A, M.B.A., Scientist, Biologics Process Development, Process Design R&D, Diagnostics Division, Abbott Laboratories

A Micro-24 bioreactor was used to determine process parameters that showed scalability in larger reactor vessels. Process parameters evaluated include pH, DO, cell density and viability, metabolite profiles and protein titer. This system provides a tool that gives predictive values at production scale, while reducing experiment time and development costs, e.g., a 24-run DOE can be performed in one experiment using a fraction of the reagents, as opposed to multiple experiments at scale.

10:00 Sponsored Presentation *(Opportunity Available – Please contact Suzanne Carroll at scarroll@healthtech.com)*

10:15 Networking Coffee Break with Exhibit and Poster Viewing

OPTIMIZING THE ENVIRONMENT

11:00 Optimizing the Culture Environment through Cell Corpse Clearance

Christopher D. Gregory, Ph.D., CSO, ImmunoSolv Limited; Professor, Inflammatory Cell Biology; Deputy Director, MRC Centre for Inflammation Research, Queen's Medical Research Institute, University of Edinburgh

Programmed cell death is a normal characteristic of all cell populations in higher organisms and the swift removal of dying cells by phagocytes is a fundamental homeostatic mechanism that underlies the health of all tissues. Cell death is also a feature of all cell cultures but because the professional phagocytes that operate so effectively *in vivo* are missing from *in vitro* systems, dead cells persist and have inhibitory effects on their viable neighbors. For this reason, cells cultured *in vitro* are sub-optimal. This presentation will consider (1) the effects dying and dead cells exert on their neighbors, (2) the mechanisms and consequences of dead-cell removal by phagocytes, and (3) the benefits – in terms of cell culture establishment, growth and productivity – of cell corpse clearance achieved in the absence of phagocytes by means of novel Dead-Cert technology.

11:30 Advantages of Dynamic 3-D Cell Culture

Rebecca Drumm, Cell Culture Scientist, KIYATEC, Inc.

Traditional cell culture for applications such as cancer research and pharmaceutical drug development has relied on a flat, 2-D monolayer of cells. Over the past 20 years, research has increasingly demonstrated that cells behave differently in 2-D versus 3-D culture. Cells exposed to dynamic flow within this 3-D *in vitro* environment respond more similarly to cells *in vivo*, when compared to a static 3-D or 2-D environment. Advantages of dynamic flow in 3-D cell culture include enhanced mass transfer affecting metabolic activity, cell-cell communication, and extracellular matrix (ECM) development. Dynamic 3-D cell culture enables cells to behave as if *in vivo*, but with the benefits and convenience of *in vitro* cell culture.

12:00pm Taking the Leap Out of Flasks into Bioreactors

John Gildea, Ph.D., Assistant Professor, Research, The University of Virginia School of Medicine

Many researchers still consider bioreactors only for large-scale or bioproduction uses. However, new bench-top and disposable systems with monitoring capabilities have transformed this niche into a growing trend for basic research and drug development/ validation activities. This talk will navigate not only through the types of bioreactors, but also the overlooked intangibles (including the use of 3-D matrices, speed and flexibility, and process workflow) when considering a purchase.

12:30 Luncheon Presentation *Sponsored by* PALL Life Sciences New Science. New Thinking.SM

Next Generation of Cell Line and Process Optimization using "High-Throughput" 10 mL Bioreactors

Tiffany D. Rau, Ph.D., Pall Corporation

Successful early stage development of cell lines and processes that will meet the demands of a commercial launch is a key factor for reducing timelines and resource demands for biopharmaceutical organizations; ultimately benefitting the patient. Cell lines and processes in the past were initially developed in uncontrolled environments that poorly modeled

bioreactors (commercial method of production) and not until late in development were cells placed in controlled bioreactor environments. Developing processes uncontrolled has risks associated with it; for instance, selecting cell lines that respond to feeds in uncontrolled environments but not under controlled conditions, thus causing additional work in cell line selection and/or process optimization. The decision to screen uncontrolled in the past was based often on resource limitations (bioreactors, number of clones, cost) not on whether it was the best way to screen or develop commercial ready products. The Micro-24™ Bioreactor is a 24 well system with individual pH, DO, and temperature control which allows users to maximize the likelihood of obtaining a "winning" cell line prior to commercialization. Data will be presented showing the Micro-24™ being utilized successfully in cell line selection activities (ranking clones) and process optimization (parameter optimization) activities and its scalability to larger bioreactors, demonstrating the advantages of a controlled "high-throughput" bioreactor system that allows rapid, very early stage process development which can contribute to shorter development timelines and lower development costs

HOW TO TREAT MAMMALIAN CELLS

1:55 Chairperson's Remarks

2:00 Cultivation of Sensitive Cell Lines – Improving Bioreactor Performance by Dynamic Membrane Aeration

Björn Frahm, Ph.D., Enzyme and Fermentation Technology, Bayer Technology Services GmbH

Although the importance of animal cell culture for the industrial (large scale) production of pharmaceutical products is continuously increasing, the sensibility of the cells towards their cultivation environment is still a challenging issue. In comparison to microbial cultures, cell cultures that are not protected by a cell wall are much more sensitive to shear stress and foam formation. Reactor design as well as the selection of 'robust' cell lines is particularly important for these circumstances. Nevertheless, even 'sensitive' cell lines are selected for certain pharmaceutical processes due to various reasons. These sensitive cell lines have even higher requirements regarding their cultivation environment. The Dynamic Membrane Aeration (DMA) bioreactor aeration is a simple concept for bubble-free aeration of such sensitive cultures. It overcomes limitations and draw-backs of previous systems. Consisting of an oscillating, centrally arranged rotor (stirrer) that is wrapped with silicone membrane tubing, it enables doubling the gas mass transfer at the same shear stress in the investigated cultivation scales of 12, 20, 100 and 200 L. Continuous cultivation at this scales allows the same product output as fed-batch cultivation does at tremendously larger reactor volumes. Apart from introducing this novel technology, the presentation comprises selected cultivation results obtained for blood coagulation factor VIII in continuous mode and a therapeutic monoclonal antibody in fed-batch mode in comparison to reference trials.

2:30 Controlled-Rate Freezing in Cryopreservation: Fact or Fancy?

Jim Moldenhauer, M.S., Senior Research Scientist, Central Cell Banking, Manufacturing Science & Technology, Eli Lilly & Company

Cells are the building blocks of biotechnology. Proper preservation of cells is paramount to the successful production of biologically-active proteins, peptides, and enzymes. Cell cultures can be considered the most critical raw material used in biological manufacturing. Consequently, it is essential that cells be preserved in a manner that ensures genetic and cellular properties remain fixed in time and space. In other words, a metabolic state of 'suspended animation' must be created in which the cells remain unchanged for many years. The most common way to induce this metabolic state is by freezing cells to very low (i.e., cryogenic) temperatures. As the rate of cooling is considered to be critical for cell survival and recovery, controlled-rate freezers (CRF) have filled a niche to serve this purpose and have, in many cases, replaced the old "styrofoam box in the -70°C freezer" method practiced for decades. However, the manufacturers of CRF do not provide any meaningful qualification or validation data to support use in a regulated laboratory environment, e.g., as governed by cGLPs or cGMPs. The user can create a custom freezing program or simply use a standard program provided by the manufacturer (more often the case). Of course, the literature (both peer-reviewed and otherwise), is replete with references to the well-accepted dogma that a cooling rate of 1°C/min. is optimal for mammalian cells. But, the questions remain, "What are the actual cooling rate(s) in your CRF"?; "How do you determine the cooling rates in your CRF"?; "How controlled is your CRF"?; "What are your options"?; and finally, "Does it really matter"? In this talk, I will attempt to answer some questions, provide insights into others, and ultimately, raise new questions for further consideration.

3:00 The Culture and Cryopreservation of Human Corneal Epithelial Stem Cells for Transplantation

Sajjad Ahmad, Ph.D., Academic Clinical Lecturer, Institute of Human Genetics and North East England Stem Cell Institute, Newcastle University

The cornea is the clear front of the eye and its epithelial surface is renewed by stem cells which can become damaged by chemical burns to the eye. Such damage results in loss of corneal clarity, persistent eye pain and blindness. Over the past few years we have developed a therapy using animal cell and product free cultured corneal epithelial stem cells to treat patients with considerable success, often restoring their vision. The challenge now is to be able to treat patients who are not able to travel to us for this stem cell therapy. This presentation will describe the results from the next phase of our research, namely cryopreserving these stem cells for transportation.

3:30 Networking Refreshment Break with Exhibit and Poster Viewing

OPTIMIZING PRODUCTIVITY

4:15 Quality Attributes of Ancillary and Reagent Materials Used in Cell Manufacturing

Fouad Atouf, Ph.D., Scientist, Biologics and Biotechnology, US Pharmacopeia

Ancillary Materials (AMs), also known as Ancillary Reagents, Ancillary Products, Process Reagents, are biological and biochemical substances used in the manufacture of therapeutics that are based on or derived from cell culture (i.e., cell and gene therapies, vaccines, proteins), but are not intended to be in the final product. The quality of this type of material is critical for successful manufacturing strategies. The United States Pharmacopeial Convention (USPC) establishes standards for drugs, biologics, and excipients; but also for AMs used in manufacturing of medicinal products. USP Chapter <1043> Ancillary Materials for Cell, Gene, and Tissue Engineered Products highlights the risk assessment and mitigation strategies along with the qualification programs used for the selection of an AM to be used in cell and tissue engineering production. This presentation will discuss some of the cell culture applications used in biopharmaceutical manufacturing, and the importance of setting proper acceptance criteria and specifications for specific cell culture reagents to ensure the quality of the final product. Case studies for specific AM products will be discussed.

4:45 Manufacturing & Regulatory Issues for *Ex Vivo* Expanded Cell-Based Products

Scott R. Burger, Ph.D., Principal, Advanced Cell & Gene Therapy

5:15 Single-Batch Production of Recombinant Human Polyclonal Antibodies

Christian Müller, Ph.D., Senior Scientist, Antibody Expression, Symphogen

Symphogen has developed an expression platform, Sympress, for consistent pro-duction of target-specific recombinant human polyclonal antibodies. Simultaneously, a characterization strategy for analysis of the polyclonal products has been designed and developed, to gain regulatory acceptance of this new drug class. The first drug product manufactured by use of Sympress has now entered the clinic.

5:45 End of Conference

Cambridge Healthtech Institute's Second Annual

Affinity Tag Protein Purification

Improving Speed, Quality and Cost

August 23-24, 2010

MONDAY, AUGUST 23 - DAY ONE

12:00pm Conference Registration

1:30 Chairperson's Opening Remarks

1:40 Featured Presentation: Affinity Tags – What, How and Why?

Tag Technologies: Opportunities and Challenges in the Biopharmaceutical Industry

David Wood, Ph.D., Associate Professor, Chemical and Biomolecular Engineering, Ohio State University

Although affinity tag technologies have been ubiquitous in the laboratory for decades, they have yet to make a significant impact in large-scale protein manufacturing. This is primarily due to the requirement for tag removal from the purified product, which leads to a number of complex processing and regulatory issues. Recently, several new technologies have been developed to address these limitations, ranging from novel self-cleaving tags, to improved ligands and resins. This talk will examine the remaining obstacles to the more general adoption of these platform methods, and showcase some of the newer technologies available.

TAGS – NEW AND IMPROVED

2:20 A Closer Look at Different Protein Tags - Exploring the Pluses and Minuses

Demetrios Vavvas, M.D., Ph.D., Instructor, Ophthalmology, Massachusetts Eye & Ear Infirmary, Harvard University

2:55 Extensive Crosstalk between O-GlcNAcylation and Phosphorylation is Revealed by Novel Photocleavable Tags Combined with Electron Transfer Dissociation Mass Spectrometry

Gerald W. Hart, Ph.D., Professor & Director, Department of Biological Chemistry, Johns Hopkins University, School of Medicine

Ser(Thr)-O-linked beta-D-N-acetylglucosamine (O-GlcNAc) is an abundant and dynamic modification of nuclear and cytoplasmic proteins. O-GlcNAc regulates signaling and transcription in response to nutrients and stress (for reviews, Nature 446, 1017-1022; Am J Physiol Endocrinol Metab 295:17-28; J. Cell Sci. 123, 13-22). O-GlcNAcylation underlies glucose toxicity and impaired insulin signaling in diabetes, regulates oncogene and tumor suppressor proteins, and plays important roles in neurodegenerative disease. Progress in the study of O-GlcNAcylation has been severely hampered by difficulty in its detection and in mapping sites of attachment. Recently, chemico-enzymatic tagging of O-GlcNAc, combined with Click chemistry to attach photocleavable biotin moieties, and affinity enrichment of tagged-O-GlcNAc-peptides, have allowed highly sensitive detection and site mapping of O-GlcNAc via electron transfer dissociation (ETD) mass spectrometry (Molec Cell. Proteomics 9: 153-160; Science Signaling 3 (104), ra2). These methods have revealed that O-GlcNAc is not only surprisingly highly abundant on nuclear and cytoplasmic proteins, but also that it has an unexpected degree of extensive crosstalk or interplay with protein phosphorylation to regulate cellular processes.

3:10 Rapid Detection and Screening of HIS -tagged Proteins Using Simple, Label-Free Dip and Read Assay *Sponsored by* fortéBIO

Sriram Kumaraswamy, Ph.D., Product Manager, ForteBio, Inc.

Methods currently used for the quantitation of His-tagged proteins have limitations such as interference from other sample components, need for slow and laborious sample purification, multiple assay steps and lack of sufficient throughput. ForteBio's anti-penta HIS biosensors have been developed as a simple, one-step, Dip and Read method on the Octet label-free detection platform for the quantitation and detection of His-tagged proteins. The system is especially suited for high-throughput expression screening and the optimization of protein expression systems where sample purification needs to be avoided. The assay utilizes the Octet optical biosensing platform and involves direct, label-free detection and quantitation of his-tagged proteins that bind to highly specific penta-HIS antibody pre-immobilized on Octet biosensors. The anti-penta HIS biosensors complement other pre-fabricated and custom biosensors on the Octet platform to provide a comprehensive set of tools for use in the entire bioprocess workflow from cell line development to production monitoring and quality analysis.

3:25 Networking Refreshment Break With Exhibit and Poster Viewing

4:05 Moderated Small Group Breakout Discussions

Join an informal, facilitated discussion group, designed to discuss important and challengeing topics related to affinity tags and protein purification. This unique session allows confrerence participants to exchange ideas, brainstorm, and have an incteractive problem solving discussion. Furthermore, these breakout groups bring together scientists who share a common interest in specific topics to develop future collaborations around a focused topic.

These forums are open for discussion of scientific challenges and are not sales opportunities. We emphasize that this breakout session is for an interactive exchange amongst scientists and is not meant to be in any way a corporate or product discussion.

5:15 Breakout Group Discussion Summaries

The facilitators of the breakout group discussions will present a brief synopsis of the discussions.

5:30 Reception in Exhibit Hall

6:30 End of Day One

TUESDAY, AUGUST 24 - DAY TWO

7:30am Morning Coffee

8:25 Chairperson's Remarks

OPTIMIZING PURIFICATION

8:30 Simplified, Enhanced Protein Purification Using an Inducible, Autoprocessing Enzyme Tag

Aimee Shen, Ph.D., Scientist, Department of Pathology, Stanford University School of Medicine

We introduce a new method for purifying bacterially expressed proteins using a highly specific, inducible, self-cleaving protease tag. This method combines affinity purification, cleavage, and tag separation into a one-step purification procedure, obviating the need for exogenous proteases to remove fusion tags from target proteins. In addition, we demonstrate that this simplified procedure can enhance the expression, integrity, and solubility of intractable proteins from diverse organisms.

9:00 Efficient Trapping and Purification of Target Protein Polyclonal Antibodies from GST- Protein Immune Sera by an Epitope Affinity Column

Dan Crimmins, Ph.D., Senior Scientist, Pathology and Immunology, Division of Laboratory and Genomic Medicine, Washington University School of Medicine

Recombinant GST-proteins are widely used to produce antibodies and are also a rich source for immunoassay standards. Polyclonal immune sera from GST-proteins contain a significant amount of anti-GST antibodies. We describe a one-step epitope affinity column purification scheme that concomitantly removes these offensive GST antibodies with subsequent purification of the target protein antibodies.

9:30 Tags for Easy Detection, Purification, and Soluble Expression of Mammalian Extracellular Proteins

Junichi Takagi, Ph.D., Professor, Institute for Protein Research, Laboratory of Protein Synthesis and Expression, Osaka University, Japan

Production of proteins of extracellular origin, including receptor ectodomains, is a formidable task. Our goal is to develop cost-efficient production/purification tools applicable to projects dealing with such "difficult to express" proteins utilizing either mammalian cell or bacterial expression systems.

10:00 One-STrEP-tag for One-step Protein Interaction Analysis

Thomas Schmidt, Ph.D., COO, IBA GmbH *Sponsored by* IBA BioTAGnology

Protein:protein-interactions (PPI) are key in many biological functions and attractive targets for drug discovery.One physiological approach to assess the complexity of PPI in a living cell is co-purification of proteins expressed at their natural level with a given recombinantly expressed tagged bait protein. Based on Strep-tag® technology IBA developed the One-STrEP-tag enabling a mild and rapid purification of intact protein complexes on immobilized Strep-Tactin®. With One-STrEP-tag purities similar to two-step procedures (i.e. TAP (tandem affinity purification) are achieved in just one step for capturing also more weakly associated protein complex constituents. Furthermore the One-STrEP-tag can be used also in TAP approaches by combining a reversibly binding antibody support with Strep-Tactin. This makes One-STrEP-tag a highly versatile tool to contribute novel insights in PPI research.

10:15 Networking Coffee Break, Exhibit and Poster Viewing

11:00 Use of Alternative Affinity Methods to Purify BACE

Jeffrey Culp, Ph.D., Associate Research Fellow, Primary Pharmacology Group, Research Centers of Emphasis, Pfizer Global R & D

After refolding the catalytic domain of beta secretase (BACE), active enzyme was isolated via an immobilized inhibitor. The established method yielded BACE with high specific activity but overall recovery was poor. An alternative affinity method was developed that improved doubled yield while maintaining quality. In addition, data will be presented on the affinity purification of full length BACE which offers the challenge of a transmembrane domain.

11:30 Multitag Affinity Purification of Humain Membrane Receptors

Isabelle Mus-Veteau, Ph.D., Project Leader, Institute of Developmental Biology and Cancer, CNRS and University of Nice

Two human membrane receptors were expressed in the yeast with a multitag affinity purification sequence of 165 amino acids fused at their C-terminus allowing detection of these proteins by western blotting using antibodies directed against the hemagglutinin (HA) tag, and purification using three affinity resins (calmodulin, streptavidin, Ni-NTA).

12:00pm Highly Efficient Purification of Functional Proteins from Mammalian Cells Using HaloTag **Sponsored by:** Promega

Rachel Friedman Ohana, Ph.D., Senior Research Scientist, Promega

Although mammalian cells are preferred for producing functional mammalian proteins with appropriate post-translational modifications, purification of recombinant proteins is typically hampered by low expression levels. We have addressed this by creating a new method configured for mammalian cell culture, providing both rapid detection and efficient purification of mammalian proteins from their native environment. This approach is based on a protein fusion tag, HaloTag, which provides efficient protein purification through covalent immobilization coupled with proteolytic tag removal. HaloTag capabilities will be demonstrated through case studies of purifying multiple functional kinases from mammalian cells.

12:30 Luncheon Presentation *(Sponsorship Opportunity Available)* **or Lunch on Your Own**

1:55 Chairperson's Remarks

2:00 Histidine-Tag-Specific Optical Probes for Protein Analysis

Michael G. Fried, Ph.D., Associate Professor, Molecular and Cellular Biochemistry, Center for Structural Biology University of Kentucky

The hexahistidine (His6)/nickel(II)–nitrilotriacetic acid (Ni2+-NTA) system is widely used for affinity purification of recombinant proteins. We will describe applications of chromophoric and fluorophoric derivatives of Ni2+-NTA for the characterization of protein-protein and protein-nucleic acid interactions in a range of systems. Practical aspects such as label-binding affinities, exchange rates, and chromophoric properties will be discussed.

2:30 Speaker to be Announced

3:00 Stable Expression and Characterization of N-Terminal Tagged Recombinant BMP15

Qinglei Li, Physiology, Baylor College of Medicine

3:30 Networking Refreshment Break, Exhibit and Poster Viewing

4:15 Cleavable Affinity Tags - Lessons Learned

William Gillette, Ph.D., Senior Scientist, Protein Purification Group Leader, Protein Expression Laboratory, Advanced Technology Program, SAIC-Frederick, Inc.

Our lab makes extensive use of TEV protease for the removal of N-terminal affinity and solubility tags. I will present data on the how parameters affect this process and what we do to optimize the outcome. I will also describe our current methods for 1) evaluating the accessibility of the TEV protease recognition site and 2) predicting the success of downstream purification of the intended protein from the cleaved partner protein.

4:45 Split Fluorescent Proteins as Nearly Ideal 'Signaling Antibodies' for Protein Tagging, Capture, and Detection

Geoffrey S. Waldo, Ph.D., Team Leader, Biosciences, Los Alamos National Laboratory

Enrichment of rare or dilute proteins using high affinity tags, for example as a first step for mass spectrometry, can be accomplished by 'tandem affinity purification'. Existing antibody/epitope combinations can be expensive and bind cell components non-specifically. We have developed a suite of orthogonal epitope tags and dedicated detection proteins that are based on split fluorescent proteins. These act as 'signaling' antibodies, becoming fluorescent only upon binding the small non-perturbing epitope tag (a 15 amino acid peptide). We describe our work with split proteins from Corynactis and Aquoria that enable multiple tags to be used for multiplexed protein labeling, capture, and detection.

5:15 Expert Discussion: Cleaving – How to Optimize the Results

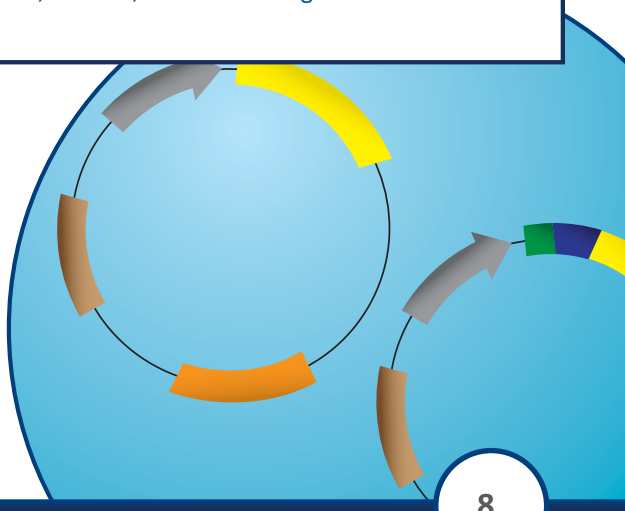
5:45 End of Conference

Reasons You Should Share Your Research Poster at The Bioprocessing Summit:

Poster Deadline is July 28, 2010

Your poster will be exposed to our international delegation

- Receive $50 off your registration
- Your poster abstract will be published in our conference materials
- Your research will be seen by leaders from top pharmaceutical, biotech, academic and government institutes



Cambridge Healthtech Institute's Second Annual

Optimizing Mammalian Cell Lines

Enhancing Expression

August 25-26, 2010

WEDNESDAY, AUGUST 25 – DAY ONE

7:30am Registration and Morning Coffee

CELL LINE DEVELOPMENT

8:20 Chairperson's Remarks *Sponsored by* cevec

Gary J. Boch, Vice President of Business Development, North America, CEVEC Pharmaceuticals

8:25 Opening Keynote Presentation:

Modifying Cellular Properties Using Microarrays – Application for Influenza Virus Production

Joseph Shiloach, Ph.D., Chief, Biotechnology Unit, National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK), NIH

The strategy of applying bioinformatics to characterize and manipulate phenotypic behaviors represents an efficient tool for altering the properties of various cell lines. Two genes, siat7e and lama4, were identified as potential targets for modifying the adhesion of HeLa cells. The effect of over-expressing sia7te was further investigated in Madin Darby canine kidney (MDCK) cells, a suitable but anchorage-dependent host for influenza virus production. Morphologically, siat7e¬-expressing MDCK cells were characterized with reduced cell-cell and cell-matrix adhesion. In growth studies, siat7e-expressing MDCK demonstrated anchorage-independent properties. In preliminary production experiments, this novel suspension MDCK cell lines supported multi-cycle replication and high-titer growth of several circulating strains of influenza viruses. This cell line is currently undergoing production evaluations to determine the feasibility of its commercial application and its possible replacement of the egg-based virus production method.

9:00 Development and Characterization of a Human Chondrocyte Cell Line That Retains Phenotype

Bryan Hoffman, Ph.D., Project Manager, Global Project Management, Morphotek, Inc.

The biology of chondrocytes is critical for understanding cartilage metabolism, however, one major limitation when studying primary chondrocytes in culture is their loss of phenotype. This talk will describe a novel approach to the establishment and characterization of human articular cartilage-derived chondrocyte cell lines. The clones were characterized by their basal gene expression profile of chondrocyte markers which resulted in selection of two clones that retained the chondrocyte phenotype. It is hoped that these cell lines which preserve important characteristics of articular chondrocytes will represent a useful model to study chondrocyte biology.

9:30 A Process for Selecting a Cell Line for Protein Products Preparing to Enter Pre-Clinical Development

Jennifer F. Nemeth, Ph.D., Principal Research Scientist and Head, Discovery Mass Spectrometry, Centocor Research and Development

10:00 Networking Coffee Break with Exhibit and Poster Viewing

TARGETED TRANSFECTION—SITE SPECIFICITY

10:45 Developing CHO Host Cell Lines Using the Artificial Chromosome Expression (ACE) Technology

Malcolm L. Kennard, Ph.D., Principal, Kennard Biologic Consultants

To improve cell line generation, the ACE System was developed based on pre-engineered artificial chromosomes with multiple recombination acceptor sites. A key feature of the ACE System is the ability to isolate and purify ACEs containing the gene(s) of interest and to transfect these ACEs with a fixed gene copy number in the same genetic environment into different host cell lines, allowing direct auditioning of these host cell lines. Results showed that there was a 5-fold difference in expression between the different CHO host cells due to cell phenotype rather than differences in gene copy number and/or location. These results demonstrate the utility of the ACE System in providing a rapid and direct technique for comparing host cell lines for optimal recombinant protein expression.

11:15 Utilization of Site-Specific Recombination (AttSite Recombinase Technology) for Cell Line Development

Susanne Corisdeo, M.S., Research Scientist, Biologics Research, Centocor R&D

We have used the AttSite® Recombinase Technology, licensed from Intrexon (RheoGene), Blacksburg, VA, for the development of mammalian production cell lines expressing biotherapeutics. This technology utilizes gene-targeting enzymes that catalyze stable and irreversible insertion of DNA at specific locations (attb sites) in the host cell genome. In the first step of this process, we developed CHO cell lines that contain the site-specific recombination point at a high-expressing site within the cell genome. Once identified, we used these attSite-containing cell lines to replace or exchange the pre-selected 'targeting sequences' with genes encoding a monoclonal antibody or other therapeutic proteins being developed by Centocor. The new genes preferentially integrated into the transcriptionally active site or 'hot spot' resulting in very consistent high-levels of protein expression. Using the AttSite™ Recombinase technology, we successfully generated candidate production cell lines with significantly less screening, which may reduce the time required for the development of production cell lines.

11:45 New Mammalian Cell Culture Applications for the Micro-24™ Microbioreactor *Sponsored by* PALL Life Sciences New Science. New Thinking.

Tiffany D. Rau, Ph.D., Pall Corporation

This workshop will demonstrate new capabilities for mammalian cell culture process development using the Micro-24™ Microbioreator system. We will illustrate the use of a new disposable bioreactor cassette for mammalian applications and describe how to rapidly implement this new technology in a cell culture process development facility. We will show how the Micro-24™ technology simplifies reactor preparation workflow, dramatically reduces set-up time (from days to minutes), enables rapid, easy feeding and sampling, and automates reactor control and data acquisition. We will provide examples of successful process optimization using a CHO expression system in the Micro-24™ Microbioreactor System.

12:00pm Presentation I: CAP and CAP-T: *Sponsored by* cevec
A Stable and Transient Expression System Based on Human Amniocytes

Jens Wölfel, Ph.D., Scientist, CEVEC Pharmaceuticals GmbH

Human CAP (CEVEC's Amniocyte Production) cells allow for stable and high yield production of recombinant proteins, with excellent biologic activity and therapeutic efficacy, as a result of authentic posttranslational modification. Based on CAP cells the transient expression system CAP-T has been developed, that enables extremely high production yields of recombinant proteins within a few days. Thus, CAP and CAP-T technologies offer the use of only one unique platform for early preclinical development through to clinical supply of recombinant biotherapeutics.

12:15 Luncheon Presentation

OPTIMIZING PROPERTIES

1:55 Chairperson's Remarks

2:00 Efficient Recombinant Cell Line Generation and Supply in a Large Compound Profiling Department

Peter Stacey, Ph.D., Principal Scientist, Primary Pharmacology Group, Pfizer, Ltd.

The reagent provision team at Pfizer's research laboratories in Sandwich, UK, provides fresh and frozen cells for multiple assays that are performed weekly, in support of ongoing drug discovery projects. Newly generated cell lines must be of a high standard in terms of assay robustness, and supply must then be maintained in either fresh or frozen format, sometimes continually for several years. The current challenges include: Increasing efficiency; succeeding with tougher targets (e.g., ion channels) and generating data with increased physiological relevance. With examples of recent data, this talk will describe the practical solutions we now have in place. These include the use of cell culture automation, viral methods for cell line generation, and improved cell sorting using L.E.A.P.

2:30 Development of a Toolbox of Human Cell Lines for Glycooptimization of Biotherapeutics

Lars Stöckl, Ph.D., Associate Director, Glycoproteins and Glycoanalytics, GLYCOTOPE GmbH

Glycosylation is one of the major post-translational modifications of biotherapeutics that depends on the cell line used for production. At Glycotope, we have generated a set of glycoengineered human cell lines to optimize the human glycosylation of biotherapeutics. Three glycooptimized products are presently in late preclinical development. By the time of the meeting, a forth one should be at the beginning of a Phase I clinical trial and a fifth one will be at a late stage Phase I clinical trial.

3:00 The Impact of Early Detection of Unstable Populations on Clone Selection and Vector Re-Engineering to Improve Cell Line Stability

Robin A. Heller-Harrison, Ph.D., Associate Director, Cell & Molecular Sciences, BioTherapeutics Pharmaceutical Sciences, Pfizer Research and Development

Successful Phase I cell line development at Pfizer relies on the ability to generate stable, high-producing clones in a short timeframe. While this objective is consistently achieved, expression instability has been observed in some clones during cell line development. The consequences of cell line instability, and the various means we have developed to detect and eliminate this instability will be presented.

3:30 Networking Refreshment Break with Exhibit and Poster Viewing

PROTEIN EXPRESSION

4:15 Optimized Transient Production of Recombinant Antibodies in HEK293 Cells for Research Applications

Thomas Schirrmann, Ph.D., Research Group Leader, Institute of Biochemistry & Biotechnology, Technical University of Braunschweig

The generation of stable mammalian cell lines is usually too time intensive and expensive for researchers that often require only milligram scales of different recombinant proteins and antibodies. To overcome this issue, we integrated transient mammalian expression in our antibody generation process. We constructed a set of mammalian expression vectors with restriction cloning sites compatible to our single chain (sc)Fv antibody gene libraries for one step subcloning into improved antibody formats. Up to 40 mg/L yield were already obtained in adherent HEK293T cells. The yield was increased to up to 200 mg/L by employing suspension adapted HEK293E cells, under serum free conditions and production in shake flasks.

4:45 Expression of Recombinant Proteins for Human Therapeutics using the PER.C6 Human Cells

John H. Chon, Ph.D., Senior Director, BioProcess R&D, PERCIVIA, LLC

Today's biopharmaceutical market requires manufacturing processes that are flexible, robust, cost-efficient, and deliver consistent product quality. PER.C6® cells have recently gained a lot of attention primarily due to the very high expression levels of recombinant monoclonal antibodies. For example, over 2 g/L of IgM and up to 13 g/L IgG production in platform fed-batch and volumetric productivity of over 27 g/L of IgG in just 2 weeks of process time using the XD® process have been reported over the past couple of years. We have successfully used this cell line to also express various recombinant proteins at high levels and with the desired quality characteristics. This presentation will describe our experiences in expressing and characterizing these recombinant proteins using the PER.C6® human cells.

5:15 Reception in the Exhibit Hall *(Sponsorship Available)*

6:30 End of Day One

THURSDAY, AUGUST 26 - DAY TWO

7:30am Morning Coffee *(Sponsor Opportunities Available)*

SCREENING CLONES

8:25 Chairperson's Remarks

8:30 Automation of Cell Line Development Workflow and Data Analysis for Increased Efficiency

Brian Majors, Ph.D., Scientist, Clinical Cellular Engineering, Biogen Idec

Cell line development traditionally requires significant hands-on laboratory time and handling massive amounts of data in an effort to identify a high-expressing cell line producing protein with the desired product quality characteristics. In an effort to increase efficiency and consistency, Biogen Idec has implemented a number of automation steps in the cell line development workflow both on the laboratory and data management sides. This presentation will discuss the Biogen Idec cell line development workflow and how we have been able to integrate automation, including liquid handling robotics, automation of assay instruments, and Microsoft Excel macros for data processing.

9:00 Rapid Development of CHEF1 Clonal Antibody Production Cell Lines

Howard Clarke, Ph.D., Principal Scientist, Process Development, CMC ICOS Biologics

(CHEF1) cell line development platform creates stable, production-quality clonal CHO cells lines in 10 weeks. In this case study, we introduce generic improvements into the Chinese Hamster Elongation Factor 1 Implementing this rapid platform. We developed three different antibody producing cell lines that show very comparable productivity and product quality to each other, and marked improvement over our previous clone development platform.Clones selected using this platform demonstrated improved productivity with further process engineering while maintaining stable expression profiles and consistent product quality, offering new insight and possible solutions for cell line development in other systems.

9:30 Screening of Antibody Expressing Clones with an Automated Deep Well System

Benjamin Wang, Ph.D., Scientist, Process Cell Culture and Fermentation, MedImmune

Large numbers of clones must be evaluated during cell line development in order to find those with the highest antibody production levels. Miniaturized systems such as multiwell plates allow many clones to be cultured simultaneously in a small footprint, but the appropriate type of plates and conditions must be chosen so that the cell growth and antibody production characteristics in wells match those in larger vessels such as shake flasks. Furthermore, automation is required for optimal and practical use of these systems for cell line development applications. In this talk, we will present our results using a fed-batch deep well culturing platform with an automated liquid handling and measurement system that was used to identify high titer monoclonal antibody expressing cell lines. Our presentation will describe the development and implementation of a powerful new platform that allows large numbers of clones to be screened and has the ability to characterize clonal diversity and test cell culture parameters relevant to large-scale bioreactor systems.

10:00 Networking Coffee Break

TRANSFECTION & ELECTROPORATION

10:45 Comparative Transfection of DNA into Primary and Transformed Mammalian Cells from Different Lineages

Dieter C. Gruenert, Ph.D., Senior Scientist, Head, Stem Cell Research Program, California Pacific Medical Center Research Institute; Professor, Dept. of Laboratory Medicine, UCSF; Professor, Dept. of Medicine, University of Vermont

Transfection efficacy of several chemical reagents to that of the Amaxa electroporation/ nucleofection system was evaluated and compared using both adherent cells and cells in suspension. With the exception of HEK 293 cell transfection, nucleofection proved to be less toxic and more efficient at effectively delivering DNA into the cells as determined by cell proliferation and GFP expression, respectively. Lipofectamine and nucleofection of HEK 293 were essentially equivalent in terms of toxicity and efficiency. Differences in efficiency and toxicity were cell type/ system specific.

11:15 Multiplexed Transposon-Mediated Stable Gene Transfer in Human Cells

Matthew H. Wilson, M.D., Ph.D., Assistant Professor, Dept. of Medicine, Nephrology Section, Michael E. DeBakey VA Medical Center, Baylor College of Medicine

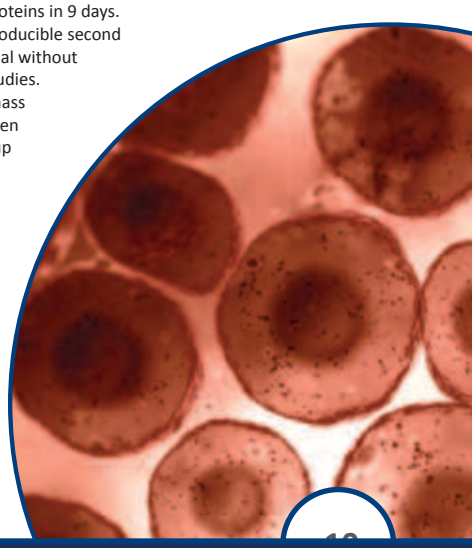
Generation of cultured mammalian cells stably expressing one or more recombinant gene sequences is a widely used approach in biomedical research, biotechnology and drug development. The piggyBac transposon system is a non-viral, plasmid based technology which offers efficient generation of mammalian cells stably expressing multiple genes. Mechanisms of gene transfer, examples of multiplexed (simultaneous multi-gene) transfer, evaluation of stability of expression long-term, and examples of application to drug-discovery will be discussed.

11:45 Towards A Fully Automated High-Throughput Phototransfection System

David J. Cappelleri, Ph.D., Assistant Professor, Mechanical Engineering, Stevens Institute of Technology

As part of a global initiative at Boehringer Ingelheim to support the rapid generation of high-quality protein for structural research, we designed and implemented a robust, automated baculoviral expression system. We adapted optimized bench-top protocols to run on the system that consistently provided multiple copies of identical small-scale biomass for up to 96 different proteins in 9 days. These methods allowed us to achieve a reproducible second passage infection in a two-day expression trial without titering the viral stock or performing MOI studies. Further downstream processing of this biomass on the automated system allowed us to screen the various constructs and triage our scale-up efforts.

12:15pm End of Conference



WEDNESDAY, AUGUST 25 – DAY ONE

7:30am Registration and Morning Coffee

BIOLOGICAL PROPERTIES

8:20 Chairperson's Remarks

8:25 Opening Keynote Presentation:

Strategies for Baculovirus Replicative Success: Viral Induction and Inhibition of Apoptosis

Paul Friesen, Ph.D., Professor, Biochemistry & Institute for Molecular Virology, University of Wisconsin-Madison

9:00 Baculovirus and Generated Proteins for Structure-Based Drug Design – More Lessons Learned

James Groarke, Ph.D., Fellow, DT Protein Structure, Novartis Institutes for BioMedical Research, Inc.

Generation of recombinant proteins in heterologous systems (specifically E.coli and Insect Cells) has become the workhorse for supplying proteins to support structure-based drug design. While these systems are generally reliable, many obstacles still remain in utilizing these expression systems to their full potential (expression levels, insolubility, scale-up issues, and post-translational modifications, for example). This talk will focus on recent advances in techniques and technologies that are being employed to overcome these aforementioned obstacles. Case Studies will be presented from the baculovirus/insect cell expression systems that highlight utilization of these technologies and improvements in the throughput process.

9:30 Insights into Baculovirus Entry and a New Highly Productive Insect Cell Line

Gary Blissard, Ph.D., Adjunct Professor, Department of Microbiology & Immunology, and Department of Entomology, Cornell University

Infection by baculovirus budded virions in cell culture is mediated by the viral envelope glycoprotein, GP64. In addition to its role in entry, GP64 is also necessary for efficient budding of progeny virions. Recent studies in my lab have focused on the mechanism of GP64-mediated viral entry, including steps of virus attachment and membrane fusion. After virus attachment at the cell surface and endocytosis, the low pH of the endosome triggers a conformational change in GP64. That conformational change drives the fusion of adjacent viral and cellular membranes. Using site directed mutagenesis of GP64 in combination with a variety of functional assays, we have addressed both structural and functional questions associated with GP64 receptor binding, membrane fusion, and virion budding. We have also recently developed a new highly productive insect cell line named Ao38, from eggs of the Black Witch Moth (Ascalapha odorata). Ao38 cells are highly permissive for AcMNPV replication and Ao38 cells produced intracellular and secreted proteins at levels exceeding those of Sf9 and High Five cell lines.

10:00 Networking Coffee Break with Exhibit and Poster Viewing

SCREENING

10:45 High-Throughput Methodology for the Rapid Screening of Gene Targets in a Baculovirus Platform

Jared Cartwright, Ph.D., Head, Protein Production Laboratory, Biology, University of York

We have developed a new set of tools for the parallel assembly of gene targets in a baculovirus platform with the subsequent rapid optimization of protein expression suitable for scale-up using bioreactor technology. The seminar will discuss all elements of baculovirus production including a rapid ligation-independent gene cloning platform suitable for the production of transfer vectors capable of intracellular and secreted protein expression. The vectors contain a copy of the green fluorescent protein (GFP) under the control of a separate promoter which serves as a marker for successful co-transfection and provides a simple mechanism to confirm viral infection. The GFP system has been further developed to provide a very simple, cheap and effective means of determining a viral titer estimation and has the advantage that it only reflects functional recombinant virus particles. We have further developed a rapid screening process to assess protein expression in suspension adapted micro-bioreactors (Applikon) and demonstrate that the parameters identified in the micro-bioreactors are readily scaled to production capacity in single-use bioreactors (scale circa 10 L). We believe that the integration of new and existing technology has produced a truly high-throughput baculovirus expression platform. Finally, we will demonstrate successful affinity purification of targets from both intracellular and secreted expression.

11:15 Screening Methods to Efficiently Produce Functional Membrane Proteins for Structural Studies using Baculovirus Expression

Ted Fox, Ph.D., Senior Director, Biology, Vertex Pharmaceuticals, Inc.

Baculovirus production has become cheaper and easier, and is readily accessible to most laboratories. For this reason the number of deposited structures of BVES-derived proteins in the Protein Data Bank continues to accelerate each year. Membrane proteins present numerous expression challenges and again, BVES continues to expand its contribution in this area. In our experience, producing membrane proteins is usually not the critical issue. However, ensuring that the protein is correctly folded and functional is essential to success in biophysical and structural efforts. Some of the most useful and robust expression and biochemical screening methods we use to generate functional membrane proteins for biochemical, biophysical and structural studies will be described.

11:45 Enhancements to Baculovirus Expression Technology *Sponsored by* BlueSky Biotech

Norm Garceau, Ph.D., CSO and President, Blue Sky Biotech

12:00pm Sponsored Presentation II *(Opportunity Available)*

12:15 Luncheon Presentation *(Sponsorship Opportunity Available)* **or Lunch on Your Own**

VACCINE DEVELOPMENT

1:55 Chairperson's Remarks

2:00 Baculovirus Expression: A Tool for Producing Immunogens for *In-Vivo* Evaluation

Indresh Srivastava, Ph.D., Associate Director, Vaccines Research, Protein Biochemistry, Novartis Vaccines & Diagnostics Inc.

2:30 Using a Novel Glyco-Engineered Insect Cell Line to Examine the Role of Glycan Structure in Influenza Virus Subunit Vaccine Efficacy

Donald Jarvis, Ph.D., Professor, Molecular Biology, University of Wyoming

We created and characterized a transgenic insect cell line capable of producing recombinant influenza hemagglutinins with more authentic N-glycans. We produced rHA in these transgenic insect cells and in the native, parental insect cell line. We then compared the relative efficacy of these two forms of rHA, with mammalian-like or insect-like N-glycans, in an aerosol challenge assay in mice with an H5N1 influenza virus. The results indicated the rHA with mammalian N-glycans was more efficacious at a lower dose than the rHA with insect N-glycans.

3:00 Utilizing the BEVS to Explore the Natural Immunity of Mosquitoes to Malaria

Richard Baxter, Ph.D., Assistant Professor, Chemistry, Yale University

Malaria is the world's most devastating parasitic disease. The search for novel malaria control strategies has led to investigation of the innate immune response of the vector Anopheles gambiae to infection by malarial parasites (genus Plasmodium). In particular, the complement-like protein thioester-containing protein 1 (TEP1) is a major factor in the

destruction of Plasmodium ookinetes. I shall describe utilization of the BEVS in addressing structural and functional questions on the mechanism of TEP1-dependent killing of Plasmodium.

3:30 Networking Refreshment Break with Exhibit and Poster Viewing

4:15 Production of a Fast-Track Influenza Vaccine using Insect Cells

Penny Post, Ph.D., VP, Quality & Regulatory Affairs, Protein Sciences Corporation

Approximately 5–20% of the US population is infected with influenza annually, where > 200,000 are hospitalized from complications and 36,000 die from flu each year. Influenza is the 6th leading cause of death among US adults. All influenza vaccines currently licensed in the US are made in embryonated eggs. Protein Sciences Corporation uses the baculovirus expression vector system (BEVS) and insect cells to produce FluBlok, a novel influenza vaccine. This production system offers a rapid response advantage for the manufacturing of an influenza vaccine, with its annual changing composition. Other advantages include rapid response to emerging strains, elimination of the need to handle live viruses (especially for pandemic influenza), and production of authentic antigen that does not require egg-adaptation. FluBlok was granted Fast Track product status by the FDA and Protein Sciences Corporation anticipates licensure of FluBlok within the 2010/2011 influenza season. This talk will describe the technology used to produce this vaccine, the clinical trial data obtained, and regulatory considerations, including our path to product licensure.

4:45 From Viral Vectors to VLPs: How the Production of AAV Vectors is Being Used as a Model

Marc Aucoin, Ph.D., Assistant Professor, Chemical Engineering, University of Waterloo

Over the past 8 years we have studied the production of adeno-associated viral vectors in insect cells following the co-infection of cells with three or more baculovirus vectors. This talk will highlight what we have learned and developed that can be applicable to the advancement of the baculovirus expression vector system for the production of VLPs and other molecules which require co-infection, including our success with low MOI high-density cultures and quantification of baculoviruses.

5:15 Reception in the Exhibit Hall *(Sponsorship Available)*

6:30 End of Day One

THURSDAY, AUGUST 26 - DAY TWO

7:30am Morning Coffee *(Sponsor Opportunities Available)*

PROTEIN EXPRESSION

8:25 Chairperson's Remarks

8:30 Parallel Protein Production in the BEVS: Processes and Reagent Tracking

Ciarán N. Cronin, Ph.D., Head, Parallel Protein Production Group, Pfizer Global R&D

A parallel protein production platform has been established at Pfizer's La Jolla, California site to support its structure-based drug design programs, and to furnish protein reagents for high-throughput screening, biophysical analysis and assay development. An integral part of this platform is the baculovirus expression vector system (BEVS). This presentation will focus on the parallel approaches used to generate protein reagents using the BEVS, including customized cloning vectors, plate-based BEVS microexpression screening, expression scale-up in Wave bioreactors, and parallel protein purification strategies. A custom in-house designed SQL database tracks all reagents from construct design through to protein deliverables, allowing colleagues global access to experimental data. The process has been applied successfully for over five years to provide purified protein reagents from multiple protein families.

9:00 Production of Recombinant Antibodies using the Baculovirus Expression System

Martine Cérutti, Ph.D., Research Director and Head, Baculovirus and Therapy Laboratory, National Scientific Research Center (CNRS)

The baculovirus insect cell system proves to be a very simple and fast tool for the expression of complex proteins such as antibodies. With the development of specific transfer vectors for the expression of heavy and light chains of antibodies, we have focused our work on optimization of amplification and cloning of cDNAs encoding VH and VL domains from human and murine B-cells. We are also interested in the glycosylation potential of lepidopteran cell lines. Even if these cells have been shown to perform most of the post-translational modifications including N- and O-glycosylations, some of the glycosyltransferases present a very low activity or are missing. New baculoviruses expressing missing glycosyltransferases such as GNT-II and GalT were constructed in order to produce galactosylated antibodies and to analyze the impact of glycosylation on their biological activities.

9:30 Efficient Protein Expression in *Bombyx mori* Larvae of the Strains Highly Sensitive to *B. mori* Nucleopolyhedrovirus

Takahiro Kusakabe, Ph.D., Associate Professor, Applied Genetics and Pest Management, Kyushu University Graduate School

By screening of silkworm bioresources, we found the highly sensitive strains to BmNPV. These strains exhibited up to about 10 times higher production of recombinant proteins than other silkworm strains tested. The silkworm strains we found can be used both for BmNPV and AcNPV systems. Simply by changing the strains, many researchers with problems in recombinant production may gain sufficient amounts of protein with the required activity.

10:00 Networking Coffee Break

INNOVATING PROTEIN EXPRESSION

10:45 BacMam Expression in Stem Cells and Primary Cells

Frederick M. Boyce, M.D., Ph.D., Assistant Professor, Neurology, Massachusetts General Hospital, Harvard Medical School

BacMam technology is the use of specialized baculoviral vectors for gene transfer into mammalian cells. BacMam vectors are widely used by industry for high-throughput compound screening and for protein expression. Earlier versions of BacMam vectors achieved efficient expression in a limited number of cell types. In this talk we will review recent advances in BacMam vectors which allow efficient expression in a wide variety of cell types including primary cells and stem cells.

11:15 The MultiBac System: New Baculovirus Expression Tools for Multiprotein Complex Production

Imre Berger, Ph.D., Group Leader, Structural Biology Unit, EMBL Grenoble

Most eukaryotic proteins exist as large multicomponent assemblies with many subunits, which act in concert to catalyze specific cellular activities. Many of these are only present in low amounts in their native hosts, impeding purification from source material. Unraveling their structure and function at high resolution will often depend on heterologous overproduction. The baculovirus/insect cell expression system is particularly useful for this purpose, for many reasons. We have developed MultiBac, a BEVS designed for multiprotein complex expression. Recombinant expression of multiprotein complexes for structural studies can entail considerable, sometimes inhibitory, investment in both labor and materials, in particular if altering and diversifying of the individual subunits are necessary for successful structure determination. Our laboratory has addressed this challenge by developing technologies that streamline the complex production and diversification. We have addressed parallelization and automation of gene assembly for multiprotein complex expression by developing robotic routines for multigene vector generation. Our MultiBac technology and several of its successful applications will be presented.

11:45 Automating Baculovirus Expression Screening

Alycia Shoultz, M.S., Scientist IV, High-Throughput Biology, Boehringer Ingelheim Pharmaceuticals, Inc.

12:15pm End of Conference

THE BIOPROCESSING SUMMIT

Cambridge Healthtech Institute's Second Annual

Practical Solutions for Today's Laboratory Challenges

August 23-26, 2010 • Marriott Long Wharf Hotel, Boston, MA

Key Code 1081 F

REGISTRATION INFORMATION

❑Mr. ❑Ms. ❑Mrs. ❑Dr. ❑Prof. Name _____

Job Title _____ Div./Dept. _____

Company _____

Address _____

City/State/Postal Code _____

Country _____

Telephone _____

How would you prefer to receive notices from CHI? Email: ❑Yes ❑No Fax: ❑Yes ❑No

Email* _____ Fax _____

*Email is not a mandatory field. However, by excluding your email you will not receive notification about online access to pre-conference presenter materials, conference updates, networking opportunities and requested eNewsletters.

SUMMIT PRICING - BEST VALUE!

(Includes access to 2 conferences, excludes short courses)	Commercial	Academic, Government, Hospital-affiliated
Early Registration Discount until June 4, 2010	❑ $1845	❑ $795
Advance Registration Discount until July 23, 2010	❑ $1995	❑ $875
Registrations after July 23, 2010 and on-site	❑ $2195	❑ $945

REQUIRED – Please select the two conferences you're most likely to attend :

August 23-24 (Choose One)
- ❑ Affinity Tag Protein Purification
- ❑ Optimizing Cell Culture Technology

August 25-26 (Choose One)
- ❑ Baculovirus Technology
- ❑ Optimizing Mammalian Cell Lines

SINGLE CONFERENCE PRICING

(Includes access to 1 conference, excludes short courses)	Commercial	Academic, Government, Hospital-affiliated
Early Registration Discount until June 4, 2010	❑ $1245	❑ $475
Advance Registration Discount until July 23, 2010	❑ $1395	❑ $545
Registrations after July 23, 2010, and on-site	❑ $1595	❑ $625

REQUIRED – Please select the one conference you're most likely to attend :

August 23-24
- ❑ Affinity Tag Protein Purification
- ❑ Optimizing Cell Culture Technology

August 25-26
- ❑ Baculovirus Technology
- ❑ Optimizing Mammalian Cell Lines

SHORT COURSE PRICING (Includes access to short course only)

Single Short Course	❑ $695	❑ $395
Two Short Courses	❑ $995	❑ $695

Please select the short course(s) you will attend:

Monday, August 23 (Choose One)
- ❑ Optimizing Media – *Achieving Super Soup*
- ❑ Affinity Tag Purification Systems

Tuesday, August 24
- ❑ *E.coli* Innovations

POSTER DISCOUNT

❑ $50 off ❑ $50 off

❑ I cannot attend but would like to purchase The Bioprocessing Summit conference CD for $750 (plus shipping). Massachusetts delivery will include sales tax.

❑ Please send information on exhibiting and opportunities to present workshops.

PAYMENT INFORMATION

❑ Enclosed is a check or money order payable to Cambridge Healthtech Institute, drawn on a U.S. bank, in U.S. currency.

❑ Invoice me, but reserve my space with credit card information listed below.

Invoices unpaid two weeks prior to conference will be billed to credit card at full registration rate. Invoices must be paid in full and checks received by the deadline date to retain registration discount. If you plan to register on site, please check with CHI beforehand for space availibility.

❑ Please charge: ❑ AMEX (15 digits) ❑ Visa (13-16 digits) ❑ MasterCard (16 digits)

Card # _____ Exp Date _____

Cardholder _____

Signature _____

Cardholder's Address (if different from above) _____

City/State/Postal Code _____ Country _____

Please refer to the Registration Code below:

HOW TO REGISTER:

Online:
BioprocessingSummit.com

Email:
reg@healthtech.com

Phone: 781-972-5400

Fax: 781-972-5425

Group Discounts

Special rates are available for multiple attendees from the same organization. **Contact David Cunningham at 781-972-5472** to discuss your options and take advantage of the savings.

Present a Poster and Save $50!

Cambridge Healthtech Institute encourages attendees to gain further exposure by presenting their work in the poster sessions.

To secure a poster board and inclusion in the conference materials, your abstract must be submitted, approved and your registration paid in full by **July 28, 2010.** Register online, or by phone, fax or mail. Indicate that you would like to present a poster and you will receive abstract submission instructions via email.

I am interested in presenting a poster at
❒ The Bioprocessing Summit
Title
_____
_____
_____

Yes! I would like to receive a FREE eNewsletter subscription to:

❒ Weekly Update
The latest industry news, commentary and highlights from Bio•IT World

CAMBRIDGE HEALTHTECH MEDIA GROUP
chimediagroup.com

❒ eCliniqua
Innovative management in clinical trials

CHI Insight Pharma Reports
A series of diverse reports designed to keep life science professionals informed of the salient trends in pharmaceutical technology, business, clinical development, and therapeutic disease markets. For a detailed list of reports, visit InsightPharmaReports.com, or contact Rose LaRaia at rlaraia@healthtech.com, 781-972-5444.

Barnett Educational Services
Barnett is a recognized leader in clinical education, training, and reference guides for life science professionals involved in the drug development process. For more information, visit www.barnettinternational.com.

Additional Registration Details

Each registration includes all conference sessions, posters and exhibits, food functions, and a copy of the conference proceedings link.

Register 3 - 4th is Free! Individuals must register for the same conference or conference combination and submit completed registration forms together for discount to apply. Please reproduce this registration form as needed.

Group Discounts are Available! Special rates are available for multiple attendees from the same organization. For more information on group discounts contact David Cunningham at 781-972-5472

Handicapped Equal Access
In accordance with the ADA, Cambridge Healthtech Institute is pleased to arrange special accommodations for attendees with special needs. All requests for such assistance must be submitted in writing to CHI at least 30 days prior to the start of the meeting.

Substitution/Cancellation Policy
In the event that you need to cancel a registration, you may:
- Transfer your registration to a colleague within your organization.
- Credit your registration to another Cambridge Healthtech Institute program.
- Request a refund minus a $100 processing fee per conference.
- Request a refund minus the cost ($750) of ordering a copy of the CD.

NOTE: Cancellations will only be accepted up to two weeks prior to the conference. Program and speakers are subject to change. Video and or audio recording of any kind is prohibited onsite at all CHI events.

CHI Cambridge Healthtech Institute
250 First Avenue, Suite 300, Needham, Massachusetts 02494
T: 781-972-5400 or toll-free in the U.S. 888-999-6288
F: 781-972-5425 • www.healthtech.com