8th International Bioprocessing India Conference

Recent Advancements & Applications in Bioprocessing for Biosimilars, Vaccines, and Bioenergy

Our Sponsors

Platinum Sponsor

Oxford NANOPORE Technologies™

Gold Sponsors

PALL CORPORATION

cytiva

PharmNXTbiotech — GLOBAL BY HEART | INDIAN BY SPIRIT

DYNA BIOTECH — TECHNOLOGY FOR CLIENTS

ThermoFisher SCIENTIFIC

JNC

ENZENE Biosciences Ltd.

Silver Sponsors

APPLIED MATERIALS

SERUM INSTITUTE OF INDIA — Cyrus Poonawalla Group

BECKMAN COULTER Life Sciences

nano TEMPER

HIMEDIA — FOR LIFE IS PRECIOUS

SCIEX — The Power of Precision

Amerging Technologies

Bronze Sponsors

Waters™

SPINCO

BioCloud

Agilent

Mini Sponsors

Tarsons

SETLAB

Xydor — Xydor Scientific Pvt Ltd

RAUT SCIENTIFIC

BioSphere Life Sciences Pvt Ltd

BioResource Biotech

CellBioasis

Day1, Friday, December 16, 2022

Plenary session

Prof. Abraham Lenhoff

Title: Crossing the Valley of Death: Fundamental Research in the Service of Biomanufacturing

Abstract: Discoveries in fundamental research are not always efficiently translated into applications in process development, design and manufacturing, whether in bioprocessing or in other spheres of industry and the economy. This "valley of death" is often seen as a barrier to innovation in industrial practice, and some governments have directed support specifically to overcoming these limitations. The history of the first few decades of biomanufacturing illustrates how the field has evolved to address such concerns; it reflects consistent growth in the sophistication of the methods employed in process development, much of which has included a reliance on a fundamental understanding of the sciences involved. The methods and applications involved span both upstream and downstream processing as well as experimental measurements and modeling efforts. This presentation will provide an overview of such developments, with selected case studies. The learning from past experience, most readily apparent in the development of recombinant proteins, can inform process development for emerging modalities such as cell and gene therapy.

Day1, Friday, December 16, 2022

Session 1

Title: Integrative multi-omics data driven approaches guide next generation CHO cell line and process development

Dr. Meiyappan Lakshmanan
Staff Scientist & Lead,
Bioprocessing Tech. Institute
Singapore

Abstract

Chinese hamster ovary (CHO) cells are the most prevalent mammalian cell factories for producing therapeutic biologics, due to its capacity for complex post-translational modifications, ability to grow well in suspension cultures and low susceptibility to human viral infections. Significant advances in various modules of the CHO cell line development and engineering have contributed to up to 100-fold increase in the product yields over the last three decades. In this talk, I will first present how mammalian systems biotechnology can drive rational cell line development and engineering through 3 case studies: 1) to decipher the genomic changes when a CHO cell becomes antibody producer, 2) genomic and phenotypic traits of various CHO parental cell lines and 3) characterizing product quality and titer variations upon changes in process conditions. I will then present our ongoing development of standardized compendium of CHO cells by assembling NGS data from our in-house sources as well as the ones deposited to public databases and uniformly processed it using standardized pipeline. The analysis of uniformly processed data highlights key information about the global and cell-line specific hallmarks in the transcriptomic, epigenomic and genomic landscapes of CHO cells.

Day1, Friday, December 16, 2022

Session 1

Title: Recent advances in use of *E. coli* as host for manufacturing of biotherapeutics

Dr. Dakshesh Mehta
Vice President - Technical
MJ Biopharm Pvt Ltd, Pune

Abstract

Despite several limitations, microbial host systems remain the most efficient and cost-effective for biotherapeutics production. *Escherichia coli* is often the preferred host due to ease of cloning, scale-up, high product yields, and most importantly, cost-effective cultivation. One often experiences difficulties in producing biologically active therapeutics in *E. coli* especially, which require Disulphide bond formation. Many recent advances in strain engineering with respect to expression and localisation of protein of interest, co-expression of soluble expression partners, media development and advances in fermentation process have increased the suitability of *E. coli* as host.

Day1, Friday, December 16, 2022

Session 1

Title: Upstream Processing for Recombinant Protein Therapeutics: Continuous upstream processing for maximum productivity

Dr. Sridhar Kottakota
Vice President- R&D
Biologics division of Laurus Labs

Abstract

As per the recent ICH Q13 Guideline on Continuous manufacturing (CM) of Drug substances and drug products definition, the CM involves the continuous feeding of input materials into, the transformation of in-process materials within, and the concomitant removal of output materials from a manufacturing process of biological/biotechnological entities. In the US, Food and Drug Administration (FDA) officials have been encouraging manufacturers to switch to continuous manufacturing from the more antiquated batch production process for years to improve product quality, minimize product defects and reduce shortages. There lots of advancements were taken place in the in academia and industry in continuous upstream processing for maximum productivity of Recombinant Protein Therapeutics. I am going to present a perspective overview on perfusion mode of CM in upstream processing in terms of development, application, and quality considerations.

Day1, Friday, December 16, 2022

Session 1

Title: Upstream CHO cell culture basal medium and feed: Monoclonal Antibodies and Recombinant Proteins

RAJESH KUMAR
Field Application Scientist
ThermoFisher, India

Abstract

Chinese hamster ovary (CHO) cells are the primary cellline for production of biotherapeutic proteins, accounting for up to 70% of recombinant proteins. These cells are the workhorse for bioproduction because they allow for human-like posttranslational modifications and are not susceptible to infection by human viruses. Year after year, biopharmaceutical manufacturers have successfully driven their CHO cells to deliver increased productivity. Despite these advances, the industry continues to strive toward maximizing productivity by streamlining manufacturing processes to reduce costs. To maximize productivity and reduce development time and costs, CHO workflows in the early stages of development require a higher-performing chemically defined (CD) platform with basal media and feeds optimized for specific CHO cell lines. To help provide solutions for the bioproduction industry, the Gibco™ Efficient-Pro™ Medium and Gibco™ Efficient-Pro™ Feeds1 and 2 were developed using a traditional and multiomics modeling approach to improve recombinant protein production in CHO cells. The CD Efficient-Pro Medium is supplemented with either of the corresponding single-part feeds designed for optimal performance with specific CHO cell lines. Efficient-Pro Feed 1 is designed primarily for CHO-K1 cells and Efficient-Pro Feed 2 for CHO-S and DG44 cells.
The performance of Efficient-Pro Medium and Feeds was evaluated using CHO-K1, CHO-S, and DG44 cells and compared to another supplier's commercially available basal medium and 2-part feed. The cell lines were evaluated in a 14-day fed-batch IgG productivity study with cell growth, viability, titer, and specific productivity assessed at multiple time points.

Day1, Friday, December 16, 2022

Session 2

Title: Impact of physicochemical and Biochemical Characterization on bioprocess development for manufacturing of recombinant bio therapeutics protein

Dr. Sourav Majumdar
Manager, Serum Institute of India Pvt. Ltd, Pune

Abstract

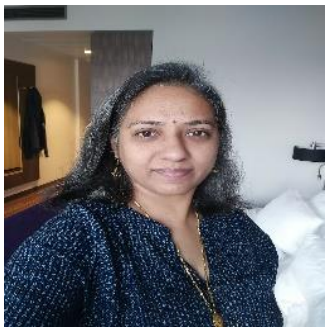
Assessment of physicochemical characterization of recombinant biotherapeutic is essential for bioprocess development since the biological system in which such a protein product is produced can have a significant effect on the structure and function of the product itself. In recent market scenario, manufacturer and regulatory agencies both are facing challenges for evaluation of productive bioprocess development without support of characterization study on therapeutics. The complexities of the recombinant biotherapeutics molecule aim to cover the primary, secondary and tertiary structure that insights into the critical quality attributes through characterization study. Large size and complex nature of Recombinant proteins are required ultramodern analytical and functional characterization to define their critical quality attributes as well as any chemical modification during bioprocess. The study was focussed on importance of characterization in bioprocess development. In general, Primary level characterization was emphasised on structural information of protein backbone; whereas Secondary, tertiary and quaternary structure is often collectively termed as the higher order structure (HOS) of a protein and demonstrated the correct folding and three-dimensional shape of a biomolecule.

Nevertheless, the challenges in physicochemical characterization are still questionable to address bioterapeutic development at different stage. Moreover, PTM of recombinant protein therapeutics was addressed by using multidimensional bioanalytical techniques to define critical quality attributes (potency and immugenicity) of the molecule. Further, bioanalytical characterization, will improve the yield of the product with safety and efficacy. Hence Biophysical characterization is attested as a promising approach to achieve a successful bioprocess.

Day1, Friday, December 16, 2022

Session 2

Title: Multiparameter Stability Characterization of Early-Stage Formulations in Characterization of Biologics



Saji Menon
Senior Application Scientist
NanoTemper Technologies

Abstract:

Biologics are an increasingly important therapeutic modality and characterization of biologics and the associated workflows from early discovery to final formulation can often be very complex, time-consuming and lack accuracy and precision needed to appropriately monitor drug candidates.

Here we demonstrate Prometheus Panta that measures thermal unfolding, particle sizing, molecular weight of aggregates & aggregation simultaneously throughout the entire thermal ramp which gives high-quality stability results to select the efficient drug targets in biologics. It predicts biologics developability profiles through early-stage screening and comparability studies to validate that changes to manufacturing processes or sites don't affect the drug product. Panta uncovers undesirable physical and chemical properties that impact solubility, stability, aggregation etc. Full antibody molecules can be engineered with higher thermodynamic stability and more efficient folding in mind to determine which ones move forward to the next stage of drug development to evaluate stability attributes in Pre formulation and Formulation studies. Finally, comparability studies that take place when changes in the manufacturing process are introduced, to make sure they won't negatively impact the quality, safety, and efficacy of the drug. For this analysis, stability is tested pre- and post-change.

Hundreds of samples can be measured per day, enabling stability screening with an unprecedented throughput and accuracy. This platform provides robust, efficient, and accurate analysis and characterization of biologics.

Day1, Friday, December 16, 2022

Session 2

Title: Sialic Acid Analysis made easy!

Sunil B Lakhmapure

Technical support Specialist
Agilent Technologies Pvt Ltd

Abstract

Glycans are carbohydrates composed of monosaccharides arranged into many different possible oligosaccharide structures based on composition and linkage position. Sialic acid capping at the non-reducing terminal of N- or O-glycans can serve a key role in mediating the effectiveness of therapeutic glycoproteins. Depending on the molecule and the application, terminal sialic acid may reduce the rate of clearance, reduce antibody-dependent cellular cytotoxicity (ADCC) activity, or can be anti-inflammatory. Two common sialic acid species found in biotherapeutics are N-acetylneuraminic acid (NANA or Neu5Ac) and N-glycoylneuraminic acid (NGNA or Neu5Gc). Neu5Ac is generally the predominant species while Neu5Gc is not synthesized by humans and its presence on biotherapeutics can potentially be immunogenic. Therefore, it is essential to monitor both the absolute quantity of sialic acid and the levels of different sialic acid species present in therapeutic glycoproteins.
This talk discusses new and improved workflows for the preparation, separation, and detection of sialic acids.

Day1, Friday, December 16, 2022

Session 2

Title: Solution NMR methods for bio-similarity assessment of peptide therapeutics

Aswani K. Kancherla
Research Scientist
ICT, Mumbai.

Abstract

Establishing comprehensive structural similarity is one of the critical criteria for approval of biosimilar therapeutic products. Solution Nuclear Magnetic Resonance (NMR) spectroscopy is a powerful method that can provide unambiguous information about primary, secondary, tertiary, and quaternary (higher order) structural information of peptide therapeutics. The presentation will introduce NMR spectral parameters and describe the features and interpretation of routinely acquired homonuclear (2D- COSY, TOCSY, and NOESY/ROESY) and heteronuclear (2D- HC-HSQC and HN-HSQC) spectra for obtaining complete sequence specific assignments. Such NMR spectra for which sequence specific assignments are completed form the foundation for a thorough structural analysis. Examples from on-going studies in our group will be used to illustrate the utility of assigned NMR spectra in obtaining information about secondary structure, conformation of the X-Pro peptide bonds, post-translational modifications such as disulphide bond formation and C-terminal amidation, and utility of diffusion ordered NMR spectroscopy (DOSY) for measuring translational diffusion coefficients to probe higher order structures in therapeutic peptides. NMR studies to establish bio-similarity often encounter challenges in the form of low concentration of the active molecule and interference from excipients used in the formulation. The presentation will discuss the work-flow and sample preparation approaches that we are employing in our group to overcome such challenges.

Day1, Friday, December 16, 2022

Session 3

Title: Alternate lignocellulolytic enzymes for bio-refineries – new organisms and processes

Dr Rajeev Sukumaran
Senior Principal Scientist,
NIIST , Thiruvananthapuram

Abstract

Lignocellulose (Plant biomass) is considered as one of the most feasible and renewable sources of energy, chemicals and materials for sustainable development. The sugar platform for biomass conversion to bioethanol, chemicals and other value added products require lignocellulose breakdown, achieved most efficiently through enzymatic hydrolysis. Enzymes are the most critical components in biorefineries and the major contributor to the operational costs. Cost reduction of enzyme involves improving the productivity of cellulase producing organisms, overcoming their tight regulation control, creation of efficient enzyme blends etc., and involves significant R&D efforts both on its bioprocessing and the molecular biology of fungi, which are currently the best producers of biomass hydrolyzing enzymes. While most of the existing biomass hydrolysing enzymes in the market are derived from variants of T. reesei RUT-C30, considered as the work horse for cellulolytic enzyme production, alternate sources of the enzyme are essential, as *T. reesei* enzymes may not be efficient in hydrolysis of all feedstock and also due to its inherent limitation of having low beta-glucosidase titres, the enzyme being the rate limiting one in cellulolytic. CSIR has been developing enzymes for biomass hydrolysis based on indigenous fungi including P. janthinellum and Aspergillus species, the former for cellulases and later for. Beta glucosidase production. Cocktails of these enzymes were demonstrated to be efficient in hydrolysis of rice-straw and other agro-residues and often at par with some of the best enzymes in use today in biorefineries. Genomic and proteomic studies of these fungi have revealed interesting features like a higher number of CAZymes and multiplicity of key enzymes involved in hydrolysis. Besides known regulators of cellulase expression, P.janthinellum was also found to harbour previously undescribed transcription factors.

Day1, Friday, December 16, 2022

Session 3

Title: Development of Biocatalytic Process for the Production of 6-Aminopenicillanic Acid Using Penicillin V Acylase for Synthetic Antibiotics

Dr.V.Koteswara Rao,
Senior Scientist,
CSIR-NCL

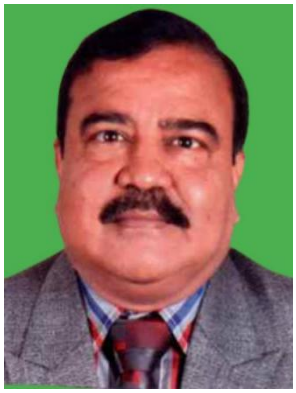
Abstract:

Penicillin V acylase (PVA, EC 3.5.1.11) hydrolyzes the side chain of phenoxymethylpenicillin (Pen V) and finds application in the manufacture of active pharmaceutical intermediate (API), 6-aminopenicillanic acid (6-APA) used in semisynthetic antibiotic production. Here, we report the scale-up of cultivation of *Escherichia coli* whole cells expressing a highly active PVA from *Pectobacterium atrosepticum* and their encapsulation in polyvinyl alcohol−poly (ethylene glycol) Lentikats hydrogels. A biocatalytic process for the hydrolysis of 2% (w/v) Pen V was set up in a 2 L reactor using the Lentikats-immobilized whole cells, with a customized setup to enable continuous downstream processing (CDSP) of the reaction products. The biocatalytic reaction afforded complete conversion of Pen V for 10 reaction cycles, with an overall 90% conversion up to 50 cycles. The bioprocess was further scaled up to the pilot-scale at 10 L, enabling complete conversion of Pen V to 6-APA for 10 cycles. The 6-APA and phenoxy acetic acid (POA) products were recovered from downstream processing with isolated yields of 85−90 and 87−92%, respectively. Immobilization in Lentikats beads improved the stability of the whole cells on storage, maintaining 90−100% activity and similar conversion efficiency after 3 months at 4°C. The robust PVA biocatalyst can be employed in a continuous process to provide a sustainable route for bulk 6-APA production from Pen V.

Day1, Friday, December 16, 2022

Session 3

Title: Bioenergy:-Fuel for India & SV BIOTECH development



Dr. Rajiv Lochan
Director
SV Biotech, Pune

Abstract

Natural resource depletion is a crucial environmental problem facing the country. Fossil fuel consumption ends up in the emission of greenhouse gases during power generation, which is responsible for global warming and climate change. The energy demands in India are increasing relatively at a high rate due to the increasing population, living standard, and economic development. The consumption of energy is relatively more than the generation of energy. India has limited resources like fossil fuels, which will soon be exhausted. All over the world, people are making efforts to shift to renewable sources of energy from Biofuels. To satisfy the endless energy demands, India is also moving towards an alternate energy source, renewable energy. Our country has good potential for developing solar, wind, hydropower, biomass, and biogas energy. This paper reviews the renewable energy scenario of India's availability of Biofuels as a renewable energy potential of India.

SV Biotech developed a patented fermentation technology, advance modified &patented media ,perfusion technology, distillation technology, and technology process for biofuel generation. Our various innovative technologies and adopted processes resulted in very high and reliable improved Ethanol production performance compared to conventional technology, as we will be contributing with hand in hand of our Government to reduce the petroleum requirement by blending of 20 % ethanol .

Day1, Friday, December 16, 2022

Session 4

Title: Platform approach for efficient AAV purification

Dr. Sandip Namdev Kadam
Thermofisher, India

Abstract

Cell and gene therapy have been rapidly growing and provides real solutions to many previously difficult to treat diseases. The FDA has predicted 10–20 new cell and gene therapies will be approved annually by 2025.

The interest in gene transduction using viral vector for gene therapies has continued to rise. Adeno-associated virus (AAV) vectors are the leading platform for gene delivery for the treatment of a variety of human diseases and has been approved for treating inherited blindness and spinal muscular atrophy.

With the increased in number of clinical successes for AAV gene therapy, a need exists for scalable commercial production platform to ensure manufacturing of AAV vectors with optimal purity, potency, and consistency. The large-scale production of the AAV vectors includes upstream and downstream processing. Upstream processing includes cell culture, transfection, and harvest. While, downstream processing includes cell lysis, clarification and separation AAV vectors.

Chromatographic purification remains challenging due to low AAV titer and presence of large number of process/product related impurities. Downstream challenges can be overcome by using the POROS™ CaptureSelect™ AAVX Affinity Resin. It has PSDVB as a backbone, POROS, which facilitates higher operating flow rates and mass transfer.

POROS™ CaptureSelect™ AAVX resin has been proven as a platform approach for set of AAV serotypes that includes AAV1 to AAV9, AAVrh10 and synthetic serotypes. It was demonstrated to have high static and dynamic binding capacity for a broad range of natural and synthetic serotypes tested. The resin implementation in process reduces the number of steps and hence improved the overall process yield.

Day1, Friday, December 16, 2022

Session 4

Prof. Abraham Lenhoff

Title: Mechanisms of Persistence of Host-Cell Proteins in Bioprocessing

Abstract: For protein therapeutics produced using recombinant DNA technology, the many proteins native to the producing host cells represent impurities that must be removed. These host-cell proteins (HCPs) have long been monitored collectively but in recent years an additional focus has been on small amounts of individual proteins that can be deleterious to the product or the patient; such HCPs have sometimes been found to persist into the final drug product. This presentation will discuss the mechanisms suspected of causing HCP persistence and efforts to improve the reliability of HCP removal. These include instances of HCP binding to the product, akin to hitch-hiking, as well as phenomena that can contribute to the formation of aggregates that contain both the product and the HCP impurities.

Day1, Friday, December 16, 2022

Session 5

Title: Unlocking vaccine & viral vector manufacturing potential with novel bioprocessing technology

Ravindra Patel, Founder
OmniBRx Biotechnologies Pvt Ltd.
Gujarat, India

Abstract

Presenting the LIMITLESS BIOPROCESSING approach towards intensified production of Vaccines, Viral vectors, stem cell therapies and other biologics. A Novel single-use bioreactor platform has been evolved using a proprietary Dynamic-Bed technology for large-scale Vaccine production and viral vector manufacturing. The CellBRx single-use bioreactors provide ultimate process scalability and significant cost reduction during vaccine manufacturing process. It's a fully automated & robust platform for vaccine manufacturing with the smallest facility footprint. CellBRx bioreactors are specifically developed to reduce the batch to batch bioprocessing variability and minimizing process failures in large scale operations. CellBRx 0.5L & 5L SUBs are research and pilot scale bioreactor systems. The CellBRx 50L and 200L SUBs are production scale platforms accommodating up to 1500 m2 of surface area for cell attachment and growth which is equivalent to 17647 units of roller bottles (850 cm2 each) or 2380 units of Cell factories CF10 (6300 cm2 each). The technology has been validated by world's largest vaccine manufacturers in terms of vaccine production and high-density growth of cell lines i.e. Vero, MRC5, HEK293 and more. OmniBRx is a bioprocess engineering company providing upstream solutions to biopharma, CGT and vaccine industries. We have developed single-use bioreactor systems using an indigenously developed and patented DBR technology. Our portfolio consists of CellBRx, MiniBRx and PerfBRx bioreactor systems across multiple scales that are suitable from development to commercial scale. These bioreactors are designed to simplify different types of cell cultures viz. adherent and suspension cultures. These innovative SUBs have the smallest per liter volume footprints and are linearly scalable across all stages of the cell culture process. We provide efficient, cost-effective and high-quality disposable bioprocessing systems with hands-on technical support to help our customers solve complex cell culture challenges.

Day1, Friday, December 16, 2022

Session 6

Title: Sustainable production of food, feed, advance biomaterials and energy through Green Synthetic Biology

Dr. Santanu Dasgupta
Senior Vice President,
Reliance Industries Limited

Abstract:

Innovation in life sciences and engineering is creating opportunities to resolve the challenges human life and civilization facing today and what is upcoming in the future. Human population is growing throughout the world alarmingly resulting a continued increase in demand for food, health solution and nutrition. Existing manufacturing principles and processes is also posing huge sustainability challenges. With the onset of fourth industrial revolution, amalgamation of physical, digital and biological systems is accelerating innovations with dramatic societal and environmental impact. Synthetic biology is leading this revolution by employing living micro-organisms to produce products useful for human life and civilization in previously unthought-of markets. Manufacturers choose biology as the method of choice to efficiently produce high-performance, sustainable products and thereby, synthetic biology is at leading edge of this $4 trillion gold rush. We at RIL have developed cutting edge tools and technologies for synthetic biology to utilize the fullest potential of this opportunity. Over a decade we have developed our capability in improving photosynthetic efficiency of algae and have established a robust round-the-year algae cultivation capability. We have also made significant advances in synthetic biology, gene editing technology, bioinformatics and availability of different high-throughput technologies to increases productivity of microorganism including algae. We have leveraged this platform for making next-gen biomaterials, feed and food ingredients, and energy. Some of these developments at RIL will be discussed during the presentation.

Day1, Friday, December 16, 2022

Session 6

Title: Biocomputers with Molecular Engineered Bacteria and its applications

Dr. Sangram Bagh FRSC
Associate professor-F
SINP, Kolkata

Abstract

Biocomputing uses molecular biology parts as the hardware to implement computational devices. The implementation of synthetic genetic logic circuits in living cells paved the way for performing human designed computation by genetically engineered cells. Such cellular computations have enormous importance in biocomputer technology development at the micron scale, where microprocessor-based computers have limitation due to energy, cost and technological constrains. In this talk I will discuss our recent effort to create biocomputers with genetically engineered cells. In one work, we have created the first artificial neural networks (ANNs) with genetically engineered cells, where engineered bacteria worked as artificial 'neuro-synapses'. Here, we adapted the basic concept of artificial neural networks (ANNs) and experimentally demonstrated a broadly applicable single-layer ANN type architecture with molecular engineered bacteria to perform complex irreversible computing like multiplexing, de-multiplexing, encoding, decoding, majority functions, and reversible computing like Feynman, double Feynman and Fredkin gates. Further, adapting the idea of distributed computing and applying in the molecular biology regime, we build a biocomputer, which can solve simple maze problems. We showed an application of such biocomputers, where the engineered bacteria can silence genes in cancer cells in a logical manner. This inspires the possibility of programmed cancer gene therapy. Together, our work represents a new approach to designing and building complex cellular computation and may have significance in establishing a new platform for cellular computing, in transforming bacterial cells as ANN-enabled hardware and biotherapeutics.

Day1, Friday, December 16, 2022

Session 6

Title: Metabolic engineering of *Zymomonas mobilis* for lactic acid production

Dr. Ashish Misra
Assistant Professor
IIT, Delhi

Abstract

Zymomonas mobilis is a facultative anaerobe which is well-known for industrial-scale ethanol production. Z. mobilis has been investigated as a potential host to produce important biochemicals through metabolic engineering. However, completely diverting carbon flux away from ethanol remains a major challenge. In all product formation studies in *Z. mobilis*, high ethanol production is considered a major challenge because of the pyruvate decarboxylase gene. Unsuccessful attempts to knockout PDC for the diversion of flux necessitates to implement other metabolic strategies. In this work, we demonstrated that almost complete redirection of carbon flux can be achieved to produce lactic acid (LA). This was accomplished by expressing a suitable lactate dehydrogenase and removing other genes in the pathway and simultaneously followed by process engineering techniques. Adaptive lab evolution was done with these strains for increasing concentration of LA. But only a slight increase in LA titre and glucose uptake was observed. However, complete consumption of glucose was observed with pH-controlled fermentations. The final strain was able to consume 100 g/l of glucose producing >80 g/l of LA in batch bioreactor studies.

8th International Bioprocessing India Conference

Recent Advancements & Applications in Bioprocessing for Biosimilars, Vaccines, and Bioenergy

Our Sponsors

Platinum Sponsor

Oxford NANOPORE Technologies™

Gold Sponsors

PALL PALL CORPORATION

cytiva

PharmNXTbiotech GLOBAL BY HEART | INDIAN BY SPIRIT

DYNA BIOTECH TECHNOLOGY FOR CLIENTS

ThermoFisher SCIENTIFIC

JNC

ENZENE Biosciences Ltd.

Silver Sponsors

APPLIED MATERIALS

SERUM INSTITUTE OF INDIA Cyrus Poonawalla Group

BECKMAN COULTER Life Sciences

nano TEMPER

HIMEDIA FOR LIFE IS PRECIOUS

SCIEX The Power of Precision

Amerging Technologies

Bronze Sponsors

Waters™

SPINCO

BioCloud

Agilent

Mini Sponsors

Tarsons

SETLAB

Xydor Xydor Scientific Pvt Ltd

RAUT SCIENTIFIC

BioSphere Life Sciences Pvt Ltd

BioResource BIOTECH

CellBioasis

Day 2, Saturday, December 17, 2022

Session 7

Title: Case studies in Upstream processing of Vaccine antigens and Monoclonal Antibodies

Dr. Sambhaji Pisal
Director (R&D) and head of Patents Cell
Serum Institute of India, Pune

Abstract:

The talk highlights case studies on upstream aspects of novel bacterial / viral antigens and IND monoclonals (expressed by mammalian cell line). The desired antigens structure is vital for immune responses and vaccine efficacy in human. Similarly, the correct configuration of monoclonals is critical for affinity binding and hence effective virus neutralization. Further, the commercial viability of vaccines and immunobiologicals also depends on yields of vaccine antigens and immunobiologicals. The success of targeted production depends on the choice of host organism and expression system. The fermentation research involves establishing and maintaining high cell density as well sustained stationary phase through media and feed strategies for higher yields. This has become increasingly important with restrictions on use of animal derived media components. Serum Institute has developed world first Pentavalent (ACYWX) meningococcal conjugate vaccine containing Serogroup X polysaccharide conjugate. N. meningitidis serogroup X polysaccharide as vaccine component derived from cultivation of a novel pathogenic bacterium with intact and desired polysaccharide and maximum extraction. MenFive vaccine is recently licensed. For viral vector-based vaccine (COVID-19 Adeno virus-based Vaccines) a pre-requisite was to execute rapid and comprehensive vaccination campaigns and likewise quickly producing a sufficient number of doses for global use. The cultivation of virus on HEK cells suspension was established through optimizing the EC 50 or infectivity dose of Adenovirus viral vector that resulted in maximum antigen recovery. As on date no effective (and safe) vaccine or antiviral drugs are available for prevention or treatment of dengue. SIIPL collaborated and received CHO clone expressing anti-dengue monoclonal antibody from Visterra (USA). The CHO clone was expressing < 2 grams/lit of antibody at laboratory scale. In house researched upstream process produced monoclonal to the tune of 4.5 grams/lit. High cell densities were achieved in short time and sustaining stationary phase resulted in increased yields. This novel molecule is nearing completion of human Phase 2 trial. Dengue monoclonal was found neutralizing clinical isolates of all four dengue viruses. All the inventions are IP protected by Serum Institute of India, and shared herewith for academic purpose only.

Day 2, Saturday, December 17, 2022

Session 7

Title: Bioprocessing, fermentation technology & developments at Dyna biotech

Dr.Vinod Rangarao Patil
CMD, Dyna Biotech, Pune

Abstract

Vast formulation and process experience with antibiotics, steroid biotransformation, cell culture, vaccines, scale-up, optimization, validation and launch. Extensive experience in process design, validation in fermenters, bioreactors, upstream, downstream manufacturing and validation.Proven expertise in clean room design, equipment design, manufacturing, & validation and microbial and cell culture product setup, puts him in a niche expert group globally.

Set up his own venture **Dyna Biotech** in 2013 which is into manufacture of equipment and turnkey project execution for biotech industry Fermentation projects for major biotech companies in India, Asia Pacific region, Monoclonal antibody project for South Korea. Equipment -Fermenters, media & buffer preparation, harvest & cleaning in place systems, purification & downstream processing.Design, manufacturing, installation of fermenters, bioreactors for biotech companies in Europe, USA, South East Asia, APAC. Design, development of cGMP biotech manufacturing facility, Successful EMEA approval of Diphtheria vaccine.

Day 2, Saturday, December 17, 2022

Session 7

Title: Upstream processing for Vaccines

Mr Vasudevan Muralidaran
Cytiva, India

Abstract

Owing to the fact that Vaccines are unique to a specific disease, it brings in challenges by way of diversity in the types of cells that needs to be cultivated. No "one fits all" doesn't work here as the needs vary with individual cell line being cultivated. To bring in cohesion, the talk today would focus on recombinant vaccines being manufactured using Mammalian cell lines i.e., we would briefly cover Cell lines being used, Bioreactors adapted, microcarriers and also advancements in the field.

Day 2, Saturday, December 17, 2022

Session 7

Title: Can current available media increase vaccine productivity?

Dr. Rahul Joshi,
Business Head - Cell Biology & Immunology
HiMedia Laboratories Pvt Ltd.

Abstract:

The growing demand for vaccines has amplified the need for increased productivity and leaner production processes. Media is one of the most critical components in the vaccine production process. Not just process but selection of the right media is also important for safety, economic and regulatory reasons.
There are two ways to optimize the production process.
1) Transitioning of current classical media to serum free media
2) Optimizing existing serum free media for higher cell densities
Our product offerings as well as customized media optimization services have been specifically designed to meet this goal. Amidst the pandemic, HiMedia has worked on developing HEKin1™ and CELLin1™, both are serum-free and animal component free medium optimized for production of viral vaccines. HEKin1™ is a complete media that will support growth of HEK293 cells without further supplementation. This media has been tested for its ability to support high-density cultures of HEK293 cells in both adherent and suspension format.
CELLin1™ is a chemically defined, animal component-free medium designed for growth and maintenance of Vero, MDCK, MDBK, PK15 and MRC-5 cells. Consistent performance, scalability for use in shake flasks and bioreactors, along with simplified purification and downstream processing ensures purity and quality of the final product. We have also developed a cost-effective serum free medium for BHK cells for foot and mouth disease vaccine. HiMedia's state of art bioproduction laboratories are equipped with advanced capabilities to screen hundreds of combinations with their automated liquid handling platforms and scale them up to a 50L reactor. In conjunction, we are coming up with a world class cGMP facility which can manufacture 10 tons of media per day.

Day 2, Saturday, December 17, 2022

Session 8

Title: Next generation biosimilars: Bispecifics and ADCs

Dr. Shardul Salunke
USV, Mumbai

Abstract

Next generation biologics like Bispecific antibodies and Antibody drug conjugates (ADCs) are getting major success world over. The market value is expected to grow exponentially US$ 5.41 billion in 2021 to US$12.05 billion by 2028. Now with Indian biosimilar companies launching monoclonal antibodies in regulated market, next focus should be on manufacturing next generation biologics in coming decade. The talk here would cover the different types of next generation molecules and their specific usages. It would also cover the challenges faced in process development and in clinics.

Day 2, Saturday, December 17, 2022

Session 8

Title: PTSelect™: A novel post-transcriptional technology that enables rapid establishment of stable CHO cell lines.

Dr. Vijay Tejwani
Postdoctoral associate
State University of New York Polytechnic,
Albany, USA

Abstract

Currently, stable Chinese hamster ovary cell lines producing therapeutic, recombinant proteins are established either by antibiotic and/or metabolic selection. , PTSelect™, a novel cell line development technology utilizes a siRNA cloned upstream of the gene of interest (GOI) that is processed to produce functional PTSelect™-siRNAs, which enable cell enrichment. Cells with stably integrated GOI are selected and separated from cells without GOI by transfecting CD4/siRNA mRNA regulated by PTSelect™-siRNAs and exploiting the variable expression of CD4 on the cell surface. The presentation describes the PTSelect™ principle and compares the productivity, doubling time and stability of clones developed by PTSelect™ with conventionally developed clones.

Day 2, Saturday, December 17, 2022

Session 9

Title: Life Cycle Of A Novel IVD Product: A Case Study

Dr. Kunal Shukla (Ph.D. Biochemistry)
Principal Scientist, R&D
Advy Chemical Pvt. Ltd, Mumbai

Abstract

"No matter how fast you run, if it's in the wrong direction, you will never reach your dream."
A scientist thinks of solution to a problem at an academic or research level involving generally small scales, limited to novelty of the product and/or the process. However, there can be many different dimensions attached to this a solution, which is also true in the case of new or novel in vitro diagnostic medical devices and products. For example, one wants to introduce some novel diagnostic solution for early detection of Acute kidney injury (AKI), a medical condition which results in rapid loss of kidney function over a period of hours or few days. As per literature, there are a number of highly sensitive markers available for early detection of AKI, however, none of them are accepted in third world countries and in developing countries, majorly due to the cost involved. Among the most recent biomarkers suggested for AKI, the molecule called Neutrophil Gelatinase-Associated Lipocalin (NGAL) has been found to be suddenly up-regulated during the early stages of kidney injury. In contrast to the traditional markers like Serum Creatinine, this rapid response enables NGAL to potentially identify kidney injury at a much earlier stage. If one thinks of a diagnostic solution to be designed using this sensitive marker, then there are a number of factors to be considered. A detailed market research and validation is essential to discover the potential of NGAL. Apart from a diagnostic solution being novel, other important factors like market opportunity and potential, regulatory requirements, clinical acceptance, ease of use, affordability, etc. should also be considered while designing the product. Afterall, Designing is not about what we think works, its about what works beautifully for others.

Day 2, Saturday, December 17, 2022

Session 9

Title: Rapid advances in point-of-care testing through microfluidic technologies, in a post COVID world

Dr Dhananjay Dendukuri
CEO & Co-Founder
Achira Labs, Bengaluru

Abstract

In the post COVID world, microfluidic technologies are poised to revolutionise point-of-care medical diagnostic testing. They provide multiple benefits such as sample and reagent volume reduction, multi-parameter analysis, automated and rapid workflow and low footprint. In India, small pathology labs, doctor's offices and home testing could benefit from these technologies through instant results for infectious disease testing as well as reduce errors due to degradation or mis-labeling of samples. Further, point-of-care instruments allow for testing in doctor's office and in many remote settings which are not covered by the current centralized model, thus improving the overall health of our communities. Achira Labs has pioneered the development of an immunoassay platform using the many advantages of microfluidics. The first test panels developed include thyroid and fertility panels and have been validated against gold-standard commercial technologies. The talk will cover advances in microfluidics and point-of-care testing using Achira's technology as a case-study.

Day 2, Saturday, December 17, 2022

Session 9

Title: **Development of an efficient vitamin D3 detection strip**

Dr. Kali Kishore Reddy Tetala
Assistant Professor,
VIT, Vellore

Abstract

Vitamin D, produced in liver, is an essential pro-hormone for human health. An individual with < 50 nmol of Vitamin D are considered as Vitamin D deficient and is interlinked with several diseases such as prostate cancer, cardiovascular diseases, dementia etc. Existing detection systems have several limitations like require large sample volumes, time consuming sample preparation, requirement of large volumes of organic solvents, followed by separation and high-end instrumentation to analyse vitamin D. Also the test is very expensive and is not affordable by large Indian population.

In this regard, we have prepared a nanomaterial, characterized it and developed the nanomaterial grafted strip (NM-strip). The NM-strip was extensively investigated for its ability to detect vitamin D3 in standard samples (50 ng/mL), evaluated the Limit of Detection (LOD; using 0.1-100 ng/mL vitamin D3 concentrations) etc., and selectivity in presence of other interfering analytes such as cholesterol, vitamin A, uric acid, glucose, etc. and in human plasma.

Day 2, Saturday, December 17, 2022

Session 9

Title: Mylab Discovery Solutions: Democratizing disease diagnosis for the benefit of mankind.

Dr. Shrikant Pawar
General Manager, R&D
Mylab Discovery solutions, Pune

Abstract:

Mylab Discovery solutions have been at the forefront of the indigenization and democratization of diagnostic test kits ever since its inception. Mylab came into the limelight by launching the first RT PCR-based Covid detection kit. But what made us the first to make the kit that performed better than many other international players is to be understood. Being Asia's first and only company that placed itself among the first three to launch a NAT test that detects HIV, HCV, and HBV in blood, Mylab became a household name during the COVID era.

In the pursuit of providing solutions that are relevant to the Indian diagnostic horizon, Mylab launched the country's first POC-based rapid test kit named the Pathocatch Covid-19 Ag test followed by the much celebrated first-ever self-test kit for disease diagnostics aka the CoviSelf. We also scaled up the production capacity to more than 1 million tests a day and possess the ability to make more than 2.5 million tests per day. We also sensed the necessity of empowering the rural sector by providing them access to world-class healthcare infra at their doorstep. Mylab came up with fully equipped MyVans that catered to around 1 million samples helping reduce the diagnostic burden on the health system. We also have solutions like the Swayam platform which is a health kiosk/health ATM that is able to test approx. 70 different parameters and provide medical help on the spot. Continuing the momentum, and by harnessing all its experiences and expertise Mylab has now ventured into eradicating TB by providing an end-to-end solution required for exterminating TB from the country and subsequently from the world.

Day2, Saturday, December 17, 2022

Session 10

Title: CHO / *E.coli* : From shake flask to tonnage scale.

Dr. Sanjay Tiwari
Vice President R & D Biotech
Lupin, Pune

Abstract

Manufacturing of biologics involves culturing of cells at multiples of thousands of liter scale to meet the product demand. Such a manufacturing is usually laid up with challenges of robustness due to insufficient considerations for mixing time, oxygen transfer, carbon dioxide stripping, nutrient uptake and supply gap, control strategies for key quality attributes such as glycans, size and charge variants, hydrophobic forms, etc. An overall comprehensive assessment integrating the scale up equipment (bioreactor/ chromatography system) limitations with process control approaches is must to ensure consistent manufacturing and hence supply of drug to the market. This presentation covers suitability of usually followed scale up criteria in the industry and, also highlights need for additional quantitative/ qualitative approaches.

Day2, Saturday, December 17, 2022

Session 10

Title: Resolving Manufacturing Challenges During Scale Up and Technology Transfer – ACase Study

Vivek Farkade
General Manager – MSAT

Bharat Serums and Vaccines Ltd, Mumbai

Abstract

The timely manufacturing of biologics products depend on a productive bioprocessing workflow. It is easier to reduce risk during scale up and technology transfer when one is aware of the key production process. Building process design considering manufacturing capabilities and constraints is essential for the success of technology transfer. Early identification of large-scale requirements, data analysis and technology evaluation results into smooth technical operation and control of manufacturing process. Author will present a case study about steps taken to resolve the manufacturing challenges during scale up and technology transfer.

Day2, Saturday, December 17, 2022

Session 10

Title: Scale Up, Tech. Transfer & Manufacturing of Biopharmaceuticals

Dr. Prashant Chawla

General Manager,

Biological E. Limited, Hyderabad

Abstract:

During the production life cycle of biopharmaceuticals, the manufacturing processes always undergo technology transfer. Technology transfer is a valuable step in the developmental life cycle leading to successful commercial manufacturing. The session elaborates on Definition objective/aim, different categories of technology transfer majority of which happens intracompany or intercompany.
Many a times, the technology transfer involves scaling up of the process to get commercial benefit from the product.
The success of the technology transfer depend mostly on successful co-ordination of departments. Apart from key stakeholders i.e. Receiving unit and Sending unit, QA, QC, Analytical Development, Engineering, MSAT also play key role.

The session discusses on reason on which success and failures of the technology transfer depends.
Case study discussion on scale up strategies used during development of Diphtheria Toxoid and Typhoid Vi purified polysaccharide. Discussion involves strategies used for successful technology transfer from Manufacturing Science Scale (30L) to Commercial scale (600L). Important of Facility fit, Quality risk assessment, Development reports for upstream and downstream processes.

Day2, Saturday, December 17, 2022

Session 10

Title: Biolector XT | High Throughput Microcultivation platform for efficient Screening, Optimization and Scale-up in Bacteria, Yeast, Fungi, Insect Cells, Plant cell suspension and Algal applications.

Mr. Jagadish Bennale

Field Marketing Specialist,

Beckman Coulter Life Science

Abstract:

Reducing costs while developing efficient bioprocesses as fast as possible is a rigorous task. Intensive screenings, including a vast number of different experiments regarding strains, media composition and process conditions, have to be conducted in order to create high yields and product titers. High-throughput fermentation in microliter scale is the method of choice, as it generates high data output while cutting costs due to savings on time and workforce, automation and low material input. We introduce "Biolector XT" microbioreactor as a powerful tool for full process control in microliter scale. The BioLector XT microbioreactor is based on a standard ANSI/SLAS (SBS) microtiter plate (MTP) format, and operates with online, pre-calibrated optical sensors. Disposable 48 well MTPs enable online measurement of basic bioprocess parameters, while patented microfluidic technology supports simultaneous pH control and feeding. The microfluidic module eliminates manual liquid handling—no tubing/ pipetting required, as everything is part of the gamma-radiated ready-to-use plate. Using methods like shake flasks can increase throughput; however, little information can be collected. Also, cultivations must be paused for sampling, which can negatively affect gas transfer and culture performance.The Biolector XT can run 48 fermentations in parallel, while measuring up to six parameters at the same time, including biomass, pH, dissolved oxygen, and fluorescences, such as proteins or dyes. While measuring, the plate continues to shake, in order to avoid any pause in gas transfer to the culture. Biolector XT is an advanced tool for all of these reasons. With the geometry of the FlowerPlate and the shaking capabilities of the microbioreactor, a broad range of defined oxygen transfer rates can be achieved, permitting easy scalability to bench, pilot, and production scales.

Day2, Saturday, December 17, 2022

Session 11

Title: All You Need to Know About - Cellufine Chromatography Media to purify your vaccines: Case Studies

Mr. Manjunath Dudhanikar

Application Consultant, M R Sanghavi & Co.

Abstract:

The Cellufine™ product line offers a broad range of chromatography resins for the purification of proteins, enzymes, and biomolecules. The media based on spherical cross-linked cellulose beads, which exhibit high chemical stability / mechanical strength, higher flux and inherently bio-compatible. Applications include mAb /protein / polysaccharide purification, endotoxin removal, and used worldwide purifying vaccines, therapeutic enzymes, and virus concentration / purification. Cellulose media have significantly lower Leachables than comparable polymeric beads. Purification and recovery of case studies will be described in this presentation.

Workflow platforms include: Affinity, Gel filtration, IEX, Mixed Mode and HIC. These media resins are available for broad range of biomolecules and applications. Customized Cellufine media / ligands, and bead sizes available for challenging purification process.

For purification of virus & heparin binding protein: Evaluation of Dextran Sulfate as a Chromatography Ligand on the Surface of Cellufine™ Cellulose Beads - Introduction of New Heparin Binding Protein and Enhanced Viral Capture Resins. Cellufine™ MAX DexS is a new pseudo affinity ligand based on dextran sulfate modification. This non-animal derived affinity ligand can be used instead of immobilized Heparin. JNC offers two different types - Cellufine™ MAX DexS-HbP is mainly designed for heparin binding proteins. Cellufine™ MAX Dex-SVirS is for purifying virus and virus like particles.

For endotoxin removal: Cellufine™ ET clean is poly(ε-lysine) immobilized Cellufine. The resin binds and removes endotoxin from your sample solution.

IEX-Cellufine: A chromatography media made from the cross-linked and spherical cellulose. Each type offers excellent flow, mechanical resistance. Virtually no shrinkage and swelling is recorded under change of pH or ionic strength. Media can be autoclaved, easily regenerated and depyrogenated.

When Purity is Paramount - Cellufine™ Media Delivers!

Day2, Saturday, December 17, 2022

Session 12

Title of Talk: Bioenergy from Biomass: Sustainable Recipe to Feed an Energy-Starved Nation

Dr. Prashant Dhakephalkar

Abstract

India is a developing Nation. Obviously, there is an ever-growing demand for the energy making India the third largest consumer of the energy in world (@7%) after USA and China. Unfortunately, India remains an energy starved nation as almost 4% of its energy demand remained unmet at its peak in May-2022. Presently, most of the energy requirement is satisfied using non-renewable sources such as coal (46%), crude oil (29%), natural gas (7%), which are finite, depleting and polluting. Bioenergy from Biomass has the potential to be such viable, renewable and abundant alternative source. Lignocellulose or plant biomass is an abundant and renewable source available in the form of agricultural residue. Use of fibrolytic anaerobic fungi (FAF) may provide solution to this problem. FAF possess cellulosome, which are the multiple enzymes forming an extracellular complex. The major function of the FAF cellulosome is to efficiently degrade cellulose, the most abundant polysaccharides of plant origin in the world. Cellulosomes are comprised of several functional subunits that are known to interact with each other and with the substrate, that is cellulosic biomass. Scaffoldin glycoproteins of cellulosomes are polypeptides that are non-catalytic in nature. Cellulase and xylanase subunits integrate with a dockerins. Scafoldins contain a modules that is responsible for the adhesion of cellulose to cellulosomal concept. These properties facilitate FAF mediated degradation of cellulosome, facilitating ethanol or biogas production. Thus, augmentation of anaerobic fungi when supplemented adequately facilitate large scale production of methane from the lignocellulosic biomass. Thus, the augmentation of the start-up inoculum with FAF improves the process efficiency without any pre-treatment making the process techno-economically viable. Considering the quantum of renewable biomass available and the efficiency of the process, ‘Bioenergy from Biomass’ seems to possess potential to make Bioenergy from Biomass: as the means to satisfy the energy demand of the society

Day2, Saturday, December 17, 2022

Session 12

Title: A techno-economic analysis for production of lactic acid from bread waste using pinch technology

Prof. Sunil K. Maity

IIT, Hyderabad

Abstract:

Lactic acid (LA) is a vital platform chemical with diverse applications, especially for biodegradable polylactic acid. Bread waste (BW) is sugar-rich waste biomass generated in large quantities in residential and commercial operations. Recently, we evaluated the potential of BW for LA production by Bacillus coagulans under non-sterile conditions. This work presents a techno-economic and profitability analysis for valorizing 100 metric tons BW per day to alleviate environmental pollution with concurrent production of LA and biomethane. We compared two fermentation approaches: acid-neutral (Scenario I) and low pH (Scenario II). Traditional esterification with methanol, followed by hydrolysis of methyl lactate, was employed for downstream separation to obtain polymer-grade LA. The high-pressure steam was generated from solid debris via anaerobic digestion to complement energy demands partly. Energy consumption was further attenuated by process integration using pinch technology, with around 15% and 11% utility cost reduction for Scenario I and II, respectively. These processes were capital-intensive, with 45-48% contribution of LA production cost stemming from direct and indirect costs. Utilities were the major cost-contributing factor (21-22%) due to energy-intensive water evaporation from diluted fermentation broth. Due to additional processing steps, capital investment and operating costs were slightly higher in Scenario I than in Scenario II. LA manufacturing cost was thus somewhat more for Scenario I ($1.87/kg) than Scenario II ($1.76/kg). The minimum LA selling price was further calculated for 5-10 years payback period and 8.5-15% internal rate of return. LA was slightly more expensive for decentralized BW processing than the market price.

Day2, Saturday, December 17, 2022

Session 12

Title: Waste Fed Biorefineries – Progress and Prospects

Dr. Venkata Mohan

IICT, Hyderabad

Abstract:

Climate change, resource depletion and waste generation are major global challenges that mankind is currently facing. Waste-fed biorefineries facilitate the use of waste for the production of a variety of biobased products in a circular loop approach. Angiogenesis plays an important role in biorefineries for the transformation of waste into primary fermented products that includes platform chemicals and fuels. This communication provides an overview of biorefinery systems in the framework of the circular economy keeping waste at a focal point in the nexus of water-food-energy that can positively address decarbonization. Optimized integrations of unit operations across closed loops with process intensification in the context of resource efficiency specific to biohydrogen production will be discussed.

Day2, Saturday, December 17, 2022

Session 12

Title: Resource Autonomy / Energy Security from Food Waste

Dr. Nurial Pezarkar, Founder and CEO

NobelExcahnge Enviroment Solution Pvt. Ltd.

Abstract:

Urban Waste:
• Indian cities generate over 62 million tons of waste annually ~ 52% is food waste
• Improper disposal of food waste causes social, environmental, health and hygiene challenges
• MSW Rules 2016 suggest AD as preferred method for disposal of food waste
• Anaerobic Digestion (AD) Technology simultaneously acts as waste treatment and energy recovery process

Urban Transport:
• The transport sector contributes to 23% the total Carbon Dioxide emissions in the world (Report International Energy Agency)
• It accounts for nearly 18% of the total energy consumed in India, second only to the industrial sector
• Nearly 98% of the energy needs of transportation are met through petroleum products
• The sector is the second largest contributor to CO2 equivalent emission (CO2 e) and generates over 150 MtCO2 e.
• Within the transport sector, road transport contributes 87 per cent of the total CO2 equivalent (CO2 e) emissions

The Plan:
Create an overlap between five top priority Govt. programs viz.
¬ SATAT – CBG is the alternate fuel identified under this program
¬ Smart Cities Program – city waste to fuel provides a sustainable transport platform
¬ Swachh Bharat Abhiyaan – mandates waste to be segregated at source into dry & wet fraction
¬ Atmanirbhar Bharat Abhiyaan - CBG helps reduce dependence on imported fuel
¬ National Clean Air Program – twin climatic impact by avoidance of CHG emissions from transport sector and waste disposal
Target to set up Food Waste to CBG plants in 100 cities over next 10 years with combined capacity to process 25000 TPD food waste and producing 1250 TPD CBG & 7500 TPD Organic Manure.Achieve Carbon Emission Reduction of about 7 Million tons / year

Day 2, Saturday, December 17, 2022

Session 13

Title: Development and Manufacturing of Gene Therapy and Viral Vector Products

Dr Janet Macpherson

Business Development Manager Enterprise Solutions Cell & Gene Therapy, Cytiva, APAC

Abstract

Manufacturing methods for viral vectors have not kept pace with the clinical development of gene therapy products. Scalable and cost-effective production processes are required to manufacture safe and efficacious clinical-grade viral vectors. In this session, we present an efficient process for adenovirus-associated virus (AAV) production and scale-up in suspension cell culture, through to purified bulk product. This AAV manufacturing process was developed by evaluating and optimizing the process steps. A novel fiber technology, Fibro, addresses the downstream bottleneck at the capture step by overcoming the diffusional and flow limitations of purification using packed-bed chromatography. In addition, an analytical assay based on surface plasmon resonance was developed for AAV viral titre quantitation.

The ongoing emergence of improved upstream and downstream processes, tools, and assays, together with their optimisation will play a key role in evolving traditional AAV viral vector production into cGMP compliant gene therapy manufacturing methods at commercial scale.

Day 2, Saturday, December 17, 2022

Session 13

Title: Scalable Filtration of Recombinant Adeno-Associated Viral (rAAV) and Lentiviral (LV) Vectors

Mr. Kiran Akhade

Team Manager – Scientific Laboratory Support

Pall Corporation

Abstract

Over the past years we've witnessed the approval of several gene therapy products, and these much-anticipated therapies are beginning to deliver on their promise and to have a radical impact on patient health. Despite an increasing number of gene therapy drugs in development and clinical trials every year, this industry is still in its early stages. As the technology matures scientifically, therapies with viral vectors are advancing through clinical trials towards commercialization, bringing an increasing demand for preclinical and clinical grade viral vectors. Expansion of viral manufacturing capacity requires scalable production methods and manufacturing systems that are tailored to specific viruses. This, in turn, will facilitate the production of large quantities of virus needed to support commercial demand in full compliance with regulatory requirements. The main bottleneck for viral vector manufacturing is scalability. Downstream purification methods should be scaled to match upstream output. This usually requires large process modifications, however maintaining product quality and yield through optimized conditions at each step often remains a challenge. In this presentation we would cover:

AAV and LV downstream manufacturing platform Case studies emphasizing on filtration unit operations involved in AAV and LV downstream process specifically:

i. Clarification using depth filters, ii. Tangential flow filtration, iii Sterilizing grade filtration

Day 2, Saturday, December 17, 2022

Session 13

Title: Upstream Bioprocessing in Gene Therapy

Mr. Pankaj Salvi

Pall Corporation, India

Abstract

Gene therapy is a rapidly emerging market for biologic drug development as it offers the potential to address many currently incurable genetic diseases. With the increase in development of gene therapies and their regulatory approvals being sought, there is a pressing need for their industrial translation. Since the viral vectors have proved efficient tools for gene delivery to target cells, bringing these therapies to the patients require the meeting the dosage requirement for high quality vectors. One of the biggest challenges to this, is the scaling the bench-scale processes to commercial GMP manufacturing and managing cost of goods. Most of the technologies used in the early phase of viral vector development are bench-scale processes. Though these deliver high efficacy at small scales, many of these they are neither robust, nor useful, when scaling towards large-scale manufacturing. Hence it becomes very critical early on, to figure out the manufacturing system to implement, especially what upstream system to use. The reason for the upstream being so critical is that it is currently a major bottleneck in viral vector manufacturing. More so, for the manufacturers deciding the platform for upstream processing, whether adherent or suspension is critical to ensure they will be able to produce vectors at the right quantity, quality, and cost to support pre-clinical to clinical development, and eventually the commercial stage manufacture. In these aspects, the presentation will try to give an overview of:

-Need to match manufacturing scale to therapy demand

-Pall's scalable platforms tailored for viral vector production (AAV and Lentviral vectors)

- Suspension cell culture solution for viral vector production with Case studies

-Adherent cell culture solution for viral vector production with Case studies

Day 2, Saturday, December 17, 2022

Session 14

Title: Harnessing SCIEX solution for Biotherapeutics: *Simple Solutions to Complex Workflows*

Dr. Rohan Shah,

Sciex, India

Abstract

Introducing the SCIEX ZenoTOF 7600 system, which is powered by novel technologies: 1) the Zeno trap and 2) an alternative fragmentation by electron activation dissociation (EAD) cell and 3. Zeno SWATH. When the Zeno trap is activated, it leads to an improvement of the instrument MS/MS duty cycle, which enhances sensitivity across the entire mass range of interest. The Zeno trap improves the detection of low-level analytes, including large biomolecules and smaller molecular compounds in both untargeted information/data dependent acquisition (IDA/DDA), and targeted MRMHR workflows. In addition, Zeno SWATH DIA utilizes the Zeno trap technology in combination with SWATH DIA exclusively on the ZenoTOF 7600 system for improvements in DIA strategies. While SWATH DIA enhances, when paired with the Zeno trap technology the peptide signal is greatly enhanced because of the increased duty cycle of >90% to enable even more identification and quantification. The reagent-free and electron energy-tuneable EAD cell allows a range of different free-electron-based fragmentation mechanisms are provided within the ZenoTOF 7600 system. Examples of electron-based fragmentation include electron capture dissociation (ECD), "hot ECD" and electron impact excitation of ions from organics (EIEIO), enabling the analysis of a wide range of molecules from multiply charged to singly charged ions. **HCP:** The concentration of HCPs can span over 5 orders of magnitude at different purification stages, requiring the analytical method covers a wide dynamic range. Meanwhile, low levels of HCPs (<1 ppm) can still exist in the final DS, imposing challenges on a method's sensitivity. Using Zeno SWATH technology, the challenge for identification of low HCP can now be overcome. **Isomer:** Traditional peptide mapping workflows using CID struggle with localization of glycosylation and differentiation of amino acid isomers, such as, aspartic acid (Asp) and isoaspartic acid (isoAsp), leading to incomplete characterization of these important quality attributes. By comparison, these challenges can be addressed using Zeno EAD.**Biologic Explorer:** Along with the hardware, SCIEX brings an equivalent powerful software, which allows the user to the data analysis in much user friendly and in high-throughput process.

Day 2, Saturday, December 17, 2022

Session 14

Title: Protein Characterization Services to Support the Development of Biopharmaceutical Products

Dr. Smita Kale

Manager - Bioincubation at Venture Center (NCL Innovation Park), Pune

Abstract

Extensive analytical characterization is required at each stage of drug development- process and product development phases. It helps gain the confidence that molecule with required right structure will generate the required functional response. In general, SEC, cIEF, UV-Vis spectroscopy, CE-SDS and LC-MS techniques help in early-stage drug development and additional techniques are employed as orthogonal methods during the later stages of drug development.

During Process development, analytical tools are required to analyze the media components, leachable, characterize the different structure levels of drug substance etc. Product development involves extensive characterization of both drug substance and drug product.

The characterization strategy is majorly governed by regulatory guidelines. The developers need to refer ICH Q6B and country specific guidelines like EMEA, US-FDA, CDSCO etc. depending on the region where they plan to launch their product. At CBA, we are offering protein characterization services for all types of Biopharmaceuticals at different stages of development to help the Biopharma developers ensure the identity and quality of their product and expedite the development process. The talk will highlight the analytical tools used for characterization of biopharmaceuticals and case studies to explain how these analyses are being carried out at CBA.

Day 2, Saturday, December 17, 2022

Session 15

Title: Bioprocessing of lactic acid bacteria

Dr. Asha Kembhavi

Head, Technology Center, Cristen-Hansen, Pune

Abstract

Chr Hansen is a bioscience company and is the world's largest probiotic producer. Most of the industrially produced probiotics belong to either Lactic acid bacteria or the spore formers. Lactic acid bacteria are recognized for their fermentative ability mostly in their probiotic benefits as well as lactic acid production for various application. They are used since a long time as starter cultures to shorten fermentative process and to reduce the risk of fermentation failure in the dairy industry. In order the develop new products for new application it is necessary to optimize both the upstream and downstream processes from strain to the final product. This requires good and efficient technology to make the process robust and profitable. Lactic acid bacteria are very sensitive to lactic acid being produced which can hinder production. Lactic acid bacteria are not very stable and sensitive to oxygen, heat and moisture. Hence the production of lactic acid bacteria in an industrial set up is very challenging.
The presentation will focus on the use of new technologies to optimize the process in the shortest time to arrive at the target product profile and overcome the process challenges.

Day 2, Saturday, December 17, 2022

Session 15

Title: Landscaping of Indian gut microbiota and development of prebiotics for human health

Dr. Ashok Dubey

Senior Scientist and Lead Nutrition Sciences,

Tata Chemicals Innovation Centre, Pune, India

Abstract

Human intestine's ability to ferment a wide range of non-digestible oligosaccharides (NDOs) present in the diet is widely established. The human intestinal microbiota encode multiple critical functions impacting human health, and immune system modulation. A healthy diet rich in prebiotics that serve as a food for the good bacteria residing our gut help in maintaining healthy gut. By screening several microorganisms, we developed an efficient technology for the production of Fructo-oligosaccharides. Studies conducted with Fructo-oligosaccharides have shown that consumption of prebiotics promotes growth of good bacteria belonging to taxa Lactobacilli and Bifidobacteria. Butyrate is the favoured fuel consumed by colonocytes and emerges out essentially as the tight junction protein regulator in the gut. In a study, involving a cohort of 80, consumption of FOS increased the relative abundance of bacterial species belonging to the genera Bifidobacterium and Lactobacillus. A significant change was also observed in certain butyrate-producing microbes like Faecalibacterium, Ruminococcus and Oscillospira. FOS consumption increased the bacterial diversity and withdrawal of prebiotics consumption led to the reduction in the bacterial diversity.A pan India study involving >1000 subjects was conducted to map the Indian gut microbiota. The study was designed to understand the gut microbiota variation keeping 4 major categories viz obesity, age, physical activeness, and food habits in mind. Positive impact of FOS on butyrate-producing bacteria and FOS mediated increased bacterial diversity reinforced prebiotics role in conferring beneficial functions to the host. The talk will focus on current technologies used globally to produce prebiotics and scientific studies to decipher the interlinkages of prebiotics and gut microbiota.

Day 2, Saturday, December 17, 2022

Session 15

Title: Bioprocessing technology for Food Industry

Vivek Chaughule, Ph.D.

Head-Development & Applications,

Doehler India Pvt Ltd for WAPAC region

Abstract

Use of biotransformation technologies for value engineering in the food products.

Day 2, Saturday, December 17, 2022

Session 15

Title: Sustainable process for microalgal based nutraceuticals through biorefinery model

Dr. Saurish Bhattacharya,

CSIR-CSMCRI, Bhavnagar

Abstract

Microalgal biomass is extensively being produced all over the world for its potential application in the renewable energy, nutraceutical and biopharmaceutical sector. However, the future of the microalgal industry is reliant on expertise that would increase the biomass yield for commercial level at a lower cost. CSIR-CSMCRI has developed sustainable process for microalgal based gamma linolenic acid through biorefinery model.

A sustainable process was developed for production of γ-linolenic acid through biorefinery model wherein ε-polylysine and protein rich powder was produced as by-product using CSIR-CSMCRI's strain CSMCRI's Chlorella variabilis (ATCC PTA 12198). Microalgal mass cultivation was done in 10 open ponds each having 15m x 5m x 0.5m dimension. The total capacity of each pond will be 37 m3 wherein two ponds was kept for generating the inoculum and 8 ponds utilized for mass cultivation for generating the biomass with assumed average productivity of 30±5 g/m2/d. Further, in order to reduce the cost of the product, a viable and scalable circular economy model was designed that utilized fishery wastewater for generation of microalgal biomass through biorefinery model wherein astaxanthin along with other co-products such as ε-polylysine and polyhydroxyalkanoates as co-products were produced apart from γ-linolenic acid.

Day 2, Saturday, December 17, 2022

Session 15

Title: Engineering of a microbial host for biocatalytic synthesis of a steroid drug intermediate

Dr. Raghavendra Gaikaiwari, Hi-Tech Bio. Pune

Abstract

Engineering of a microbial host for biocatalytic synthesis of a steroid drug intermediate

Pharmaceutical companies in the steroid business seek technology for the biocatalytic production of pregnenolone, a key steroid intermediate, owing to the current multistep chemical synthesis which results in poor yield. The efforts were to develop a commercially viable technology for biocatalytic production of pregnenolone. Pregnenolone (a C-21 steroid) is biosynthetically produced by the side chain cleavage of cholesterol or phytosterol (a C-27 steroid). The reaction is catalysed by Cytochrome P450 monoxygenase enzyme complex consisting of P450 side chain cleavage (P450scc) enzyme and two redox partners viz. ferredoxin and ferredoxin reductase. Cloning and expression of cytochrome P450 enzyme complex (P450scc from the strain isolated by Hi Tech Biosciences (HTBS) and ferredoxin and ferredoxin reductase from a bacterial strain) was carried out in *E.coli* host strain. Good expression for P450scc in *E.coli* was seen and the enzyme was found to be active against phytosterol. The proof of concept was established by carrying out biotransformation of phytosterol to pregnenolone using isolated crude enzymes as well as whole cells. After optimization studies, >97% conversion of phytosterol to pregnenolone was achieved. Low solubility of sterols in the aqueous medium and possible lack of appropriate sterol uptake mechanism in *E.coli* host strain were the major problems in the technology development. In an effort to achieve the higher conversion rates and greater yield of pregnenolone, several different approaches were tried out including co-expression with AlkL genes, use of *E coli* Tol C mutant, co-expression of P450scc and its natural redox partners and finally use of the host strains which are better equipped for sterol uptake.

8th International Bioprocessing India Conference

Recent Advancements & Applications in Bioprocessing for Biosimilars, Vaccines, and Bioenergy

Our Sponsors

Platinum Sponsor

Oxford NANOPORE Technologies™

Gold Sponsors

PALL PALL CORPORATION

cytiva

PharmNXTbiotech GLOBAL BY HEART | INDIAN BY SPIRIT

DYNA BIOTECH TECHNOLOGY FOR CLIENTS

ThermoFisher SCIENTIFIC

JNC

ENZENE Biosciences Ltd.

Silver Sponsors

APPLIED MATERIALS

SERUM INSTITUTE OF INDIA Cyrus Poonawalla Group

BECKMAN COULTER Life Sciences

nano TEMPER

HIMEDIA FOR LIFE IS PRECIOUS

SCIEX The Power of Precision

Amerging Technologies

Bronze Sponsors

Waters™

SPINCO

BioCloud

Agilent

Mini Sponsors

Tarsons

SETLAB

Xydor Xydor Scientific Pvt Ltd

RAUT SCIENTIFIC

BioSphere Life Sciences Pvt Ltd

BioResource BIOTECH

CellBioasis

Day 3, Sunday, December 18, 2022

Session 16

Title: Leveraging Hybrid modeling in Bioprocess Development: Challenges and Opportunities

Dr. Vaibhav Patil
Manager of Data Science,
Asia Pacific, Sartorius

Abstract:

Cell culture processes are driven by changes in the chemical environment of the culture which elicit reactor control actions, and cell growth responses, and are sensed by signaling pathways that influence metabolism. The interactions between these forces shape the culture dynamics through different stages of cell cultivation and have a substantial effect on process productivity, product quality, and robustness. Developing a systems model that describes the interactions of these important players in the cell culture system can lead to a better understanding and optimization of processes. A few noticeable attributes of the current status of the Biopharma industry are that it depends mainly on long and costly process development timelines, Mostly wet-lab experiment-based development platforms, Large amounts of complex heterogeneous data, Key Decision making driven by data-driven modeling (i.e., MVDA or DOE).These attributes are followed by some unique challenges like complex biology not fully understood, variability and non-reproducibility of processes, pressure to adopt continuous processes, and uptake of novel therapeutics & standard workflows that don't exist yet. Hybrid modeling can bring huge value at this juncture. Hybrid modeling is a combination of data-driven & mechanistic models. These models can be further used for In-silico simulations. In-silico simulations can decrease development costs and timelines as well as standardize workflows which can decrease development complexity. I will be sharing some of our experiences while leveraging Hybrid modeling in bioprocess development at various industrial laboratories.

Day 3, Sunday, December 18, 2022

Session 16

Title: Purification of Closely Related Impurities using Multimodal Chromatography Resins – A Model Based Approach

Dr.Ganesh .T.Sivanthan
Principle Scientist,
Downstream Processing and Technology Transfer, Lupin Limited ,Pune

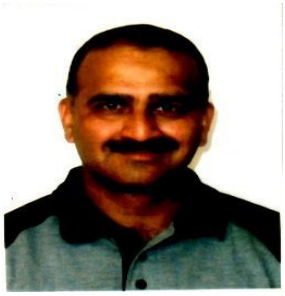
Abstract:

In recent days, the use of model - aided process development has gained popularity as a means to understand processes and products better by identifying critical and key process parameters (CKPP) and critical quality attributes (CQA) of product. This presentation will include case studies describing how the mechanistic models (classical stoichiometric displacement model and thermodynamic model) are established in the chromatography purification step to understand the selectivity in removal of the closely related clipped proteins and antibody charge variants using multimodal chromatography resins. Data will also be presented documenting that the established model predicts the process outcomes more thoroughly, providing better understanding of the separation of closely related impurities and selection of the best resin. The presentation will include the comparison and validation of trends predicted by the mechanistic model with the experimental results. The experimental results will be presented revealing how the purification outcomes coincide with the trends predicted by the model. The presentation will also highlight the key insight gained through this work and the necessity of models developed with quantifiable parameters to fit experimental observations which helps in achieving the ultimate goal of minimizing development work and forecasting process conditions simply based on protein characteristics.

Day 3, Sunday, December 18, 2022

Session 16

Title: In-Silico and Mechanistic Modeling For Bioprocesses



Mr Ravindra khare

Director of Symphony Technologies Pvt Ltd.

Abstract

Bioprocesses, like all other structured processes can be modelled in-silico. We shall explore methods of In-silico modelling, starting with mechanistic modelling, augmenting the hypothesis with data for specific processes, and building descriptive and predictive models. We shall explore the traditional regression and classification pathways of modelling and go further to explore Machine Learning and Neural Networks to build predictive in-silico models.

We shall explore the benefits and applications of in-silico modelling to speed up development and economize on resources.

Day 3, Sunday, December 18, 2022

Session 17

Title: Is That My Product Aggregate? How Subvisible Particle Identification Can Improve Your Formulations Development Workflow

Mr. Hendrick Loei,

Halo Lab, Singapore

Abstract

Subvisible particles (SVPs) in parenteral biological drugs is a safety and quality concern. Conventional subvisible particulate analysis methods such as light obscuration and flow imaging struggles with translucent aggregates or particles with refractive index very similar to that of their formulation buffer. Besides this, these methods are unable to conclusively identify the nature of the particles. The Halo Labs Aura™ system is revolutionizing SVP analysis with its high throughput, low volume, and particle identification capabilities. By combining backgrounded membrane imaging (BMI), side illumination membrane imaging (SIMI) and fluorescence membrane microscopy (FMM), the Aura™ system provided unprecedented sensitivity, ease of use and insights to the particle problems biological drug developers and manufacturers are facing. In this presentation, we will discuss how the Aura system has improved formulations development, stability programmes and process development for various biological drugs including mAbs, viral vectors, and CAR-T therapeutics.

Day 3, Sunday, December 18, 2022

Session 17

Title: Improving Protein Purification: Application of Excipients in Downstream Processing

Dr. Smita Rajput

Merk Life Science Pvt. Ltd., India

Abstract

Excipients are used to improve the stability of protein-based therapeutics by protecting the protein against a range of stress conditions such as temperature changes, pH changes, or agitation. Similar stresses are applied to proteins during downstream purification. Shifts in pH during Protein A chromatography, subsequent incubations at low pH for virus inactivation, and changes in conductivity in ion exchange chromatography can lead to aggregation, fragmentation, or other chemical modifications of the therapeutic protein. Given the potential impact on the protein's structural integrity, there is a need for approaches to reduce the risk presented by the conditions during downstream processing. For example, integration of a solution to prevent aggregation of proteins would be a more efficient strategy than implementing steps to remove multimeric forms. In this study we focused on excipients like sugars, polyols and PEG4000, which have been confirmed for their stability effects on proteins at low pH (e.g. monoclonal antibodies) using specifically designed screening assay. The objective of this work was to apply excipients in buffer systems to study their impact on several aspects during Protein A chromatography and virus inactivation. Effect of the excipients on elution patterns, stabilization of the monomer antibody, host-cell protein removal, virus inactivation rates and binding capacity of cation exchange chromatography were explored. Results of the study show that addition of excipients can have beneficial effects on Protein A chromatography and virus inactivation, without harming subsequent chromatographic steps.

Day 3, Sunday, December 18, 2022

Session 17

Title: Comparison of strategies in the development and manufacturing of low viscosity, ultra-high concentration formulation for IgG1 antibody

Dr. Vaibhav Deokar

Lupin Biotech, pune

Abstract

Present research is focused on comparative evaluation of scalable manufacturing strategies to develop ultra-high concentration (>150mg/mL at ~200mg/mL), low viscosity (<20cps) formulation for biosimilar IgG1, suitable for single, subcutaneous administration of ~600mg/3.0mL per injection. It also provides comparative evaluation of manufacturing strategies and their impact on chemical and structural stability of IgG1. The techniques used for concentration of IgG1 are tangential flow filtration (TFF), spray drying (SPD) and spray freeze drying (SFD). IgG1 was concentrated to ~200mg/mL using tangential flow filtration (TFF). Alternatively, spray dried (SPD) and spray freeze dried (SFD) IgG1 powder, was reconstituted in 30%v/v propylene glycol (PG) to form ultra-high concentration (~200mg/mL) injectable formulation.

Day 3, Sunday, December 18, 2022

Session 18

Title: Bioprocessing of natural colorant (Chlorophyll) from isolated microalgae

Dr.Kalyan Gayen,

Professor, NIT, Agartala

Abstract

Various toxic and health hazardous materials such as synthetic dyes, conventionally used as food colorant, are concerned in modern civilization. Synthetic dyes accumulated in human health, consequently causing impairment and dysfunction of various human organs. On the other hand, herbal products, medicinal or food stuffs are becoming popular these days due to their low or no side effects. The natural colorant, chlorophylls is viable alternative to solve this problem. Chlorophylls are essential compounds in sustentation of human life. They are used as additives in pharmaceuticals, cosmetic products and natural food coloring agents. In addition, dietary chlorophylls are antioxidants and anti-mutagenic substances. Hence, development of an economic production process for the chlorophylls from microalgae is relevant. North-East part of India is enormously bio-diversified and can be abundant source of microalgae for the production of commercial chlorophylls. However, the major challenges are to find out suitable microalgae that are rich in chlorophylls and also the economically viable extraction protocol. Aim of this work was to maximize the yield of chlorophylls of identified algae strain by manipulating chemical (e.g. carbon, nitrogen, phosphorus) and physiochemical parameters (pH, temperature and light intensity) and the extraction of chlorophylls in laboratory scale prior to semi-industrial scale demonstration of the operation. We investigated a cost-effective scalable process for producing chlorophylls from isolated high-yielding microalgae that will ultimately reduce the harmful effect of synthetic color. Development of end-to-end process technology for production of chlorophylls from microalgae is the novelty of the present work

Day 3, Sunday, December 18, 2022

Session 19

Title: Mechanistic insights into the stress-induced aggregation of TDP-43 in ALS

Dr. Santosh Kumar Jha
Principal Scientist and Associate Professor (AcSIR)
CSIR-National Chemical Laboratory, Pune

Abstract

Chronic environmental stress modulates the physiochemical and solvation environment of the cellular milieu and leads to the amyloid-like aggregation of proteins in neurodegenerative diseases. However, how the stress is sensed by proteins at the molecular level and consequent steps during their aggregation is not well understood for any protein. One such vital nucleic-acid binding protein is TDP-43, whose aggregation is implicated in ~97 % of cases of amyotrophic lateral sclerosis (ALS), an incurable motor-neuron disease. In this talk, I will discuss about our discovery of TDP-43 as a physiochemical stress-sensor. I will also discuss about how the amyloid-like misfolding of the protein could begin from site-specific triggers and how the energy landscapes of folding and aggregation of the functional domain of the protein is coupled by a metastable molten-globule like oligomeric form. Finally, the site-specific step-wise mechanism of the formation of amyloid-like aggregate of the protein will be discussed.

Day 3, Sunday, December 18, 2022

Session 20

Day 3, Sunday, December 18, 2022

Session 20

Mr. Manoj Dev
(ICT-Mumbai)

Title: L-Asparaginase from an acrylamide degrader, *Cupriavidus oxalaticus* ICTDB921: Production, purification and characterization

Abstract: *Cupriavidus oxalaticus*, a novel soil isolate, was found to have significant L-asparaginase (LAase) activity as compared to other screened bacterial strains. The medium and fermentation conditions were optimized for maximizing LAase production followed by its purification and biochemical characterization. The optimum conditions of pH (7), temperature (30 °C) and agitation (180 rpm), showed the highest biomass accumulation (DCW 4.76 g/L) and LAase activity (22.94 IU/mL) after 12 h at 160 mM L-asparagine. The crude LAase from this isolate showed high potency at pH 7 to 9, and temperatures from 30 to 40 °C with reasonable pH (6 to 9) and thermal (up to 50 °C) stability. The LAase was stable against most amino acids and metal ions, and it could degrade aliphatic amides like urea and formamide. The sequential purification by ammonium sulphate precipitation, thermal treatment and chromatographic separation achieved approximately 8-fold purity. The purified subunit showed a molecular weight of 78 kDa, and km and Vmax of 24.1 mM and 40 µmol/min, respectively. The study demonstrates the potential of LAase from *Cupriavidus oxalaticus* in the mitigation of asparagine in the food as well as for medicinal applications.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Nitika

IIT Delhi

Session Choice: Downstream Processing for Recombinant Protein Therapeutics

Title: Real time feedback and feedforward control of chromatographic loading and in-line concentration of monoclonal antibody fragment using Near Infrared Spectroscopy (NIR).

Abstract: Biotherapeutic molecules produced from microbial cells need refolding to achieve active structure. Most monoclonal antibody fragment (Fab) molecules produced from *E. coli* have low titer and thus need to be concentrated between refolding and chromatographic loading steps. Purification of Fab is usually a two-step process containing capture and polishing chromatography. During capture chromatography of Fabs, loading beyond a critical amount of Fab onto the column leads to non-specific binding of impurities which decreases the purity of target molecule. While loading below a certain amount would lead to decreased resin utilization. To address this issue, we have developed a Near Infrared Spectroscopy (NIR) based Process Analytical Technology (PAT) tool which combined with Partial Least Square (PLS) model can be used to determine total protein concentration of Fab molecules. The output of PLS model is feed into a python-based software which controls pump flow rate of BioSMB and in-line concentrator (ILC). In case of any deviation, the model is able to control the flow rate of ILC for the upcoming material, and flow rate of BioSMB for the existing material. This helps in better control of the process as the model is able to anticipate the changes in concentration and control unit operations before and after the surge tank using feedforward and feedback control strategies. The loading and elution profiles of various chromatographic runs are compared where deviations in total protein concentration already present in the surge tank can be observed in the loading profiles while the elution profiles remain constant. Similarly, the total protein concentration can be kept constant for the upcoming material by changing fold concentration through ILC. The constant elution profiles depict feedforward control of chromatographic loading and constant surge tank concentration depicts feedback control of ILC.

Day 3, Sunday, December 18, 2022

Session 20

Dr. Keerthiveena B
(IIT Delhi)

Session Choice: Advance Biomanufacturing capabilities: Next Generation Technology Solutions

Title: Deep Learning based pre-processing and peak identification in Liquid Chromatography

Abstract:

Proteomics strategies based on the combination of liquid chromatography and mass spectrometry (LC-MS) have been extensively employed in characterizing protein therapies at both the process and product levels. Accurate peak selection is a basic yet difficult issue in LC-MS-based omics research. Despite the fact that commercially available software packages presently provide the option of conducting peak identification and peak integration automatically, a visual examination and human adjustment of the achieved integration is still required in many instances. In this investigation, we suggested a three-step strategy for improved peak identification, which aids in analyzing chromatographic data by highlighting where peaks emerge. In the first step, autoencoder-based denoising was applied to the raw MS data in order to reduce false positives and interpret the related features of chromatographic peaks. The MS spectra are advanced to the next level based on their noise and peak score. Additionally, a model based on a convolutional neural network was constructed in order to perform automated peak identification and estimate the position and the area of the peak. The obtained results demonstrated that the proposed model is better than other state-of-the-art techniques such as Savitzky-Golay, wavelet transform and normalized least mean squares for denoising the peaks. The proposed peak detection methodology is trained and validated on real-time chromatographic samples, which is a significant benefit of using this approach.

Day 3, Sunday, December 18, 2022

Session 20

Dr. Priti Singh
(CSIR-NCL)

Session Category: Biosensing and Diagnostics

Title: Electrochemical Biosensors for the detection of Cardio Vascular Diseases

Abstract:

Electrochemical biosensor holds great promise in the biomedical area due to its enhanced specificity, sensitivity, label-free nature and cost effectiveness for rapid point-of-care detection of diseases at bedside. The working principle of electrochemical biosensor will be discussed in detail and how it can be employed in detecting biomarkers of fatal diseases like cancer, AIDS,hepatitis and cardiovascular diseases. Lastly, the discussion will be on method of fabrication of an electrochemical biosensor to detect PMPs in a blood using silica nanoparticles. Electrochemically deposited thionine doped silica nanoparticles (Th@SiNPs) and horseradish peroxidase entrapped amine functionalized mesoporous silica nanoparticles (NH 2 -MSNs) were utilized for conjugation with antibodies recognizing active confirmation of platelet glycoprotein GPIIb/IIIa and P-selectin expressed on PMPs respectively. Cyclic voltammetric analysis showed a progressive increase in reduction peak current on PMPs binding with the fabricated electrode in the presence of peroxide. The response of the sensor was linear in the range from 490 to 4080 PMPs μL −1 and could detect PMPs with a detection limit of ∼96 PMPs μL −1 . The developed sensor proved to be highly specific in identifying PMPs in plasma and offers highly biocompatible, label-free, cost-effective, ease of miniaturization, short analysis time and simple instrumentation for the detection of biomarkers for a variety of diseases. The novel sensor is expected to find widespread applications in clinical point-of-care testing.

Day 3, Sunday, December 18, 2022

Session 20

Mr. Sunil Rana
(CSIR-NCL)

Session Category: Downstream Processing for Recombinant Protein Therapeutics-II

Title: Chromatography-assisted refolding of recombinant therapeutic proteins expressed in *Escherichia coli*

Abstract:

A novel continuous refolding method was designed by direct dilution of solubilized inclusion bodies in refolding buffer using a laboratory scale tubular reactor and direct capture of peptibodies on expended bed adsorption chromatography (Protein A chromatography column). The peptibodies is an Fc fusion protein that has a fragment crystallizable region of IgG1 and a biologically active peptide fused at the C terminus and expressed in Escherichia coli in the form of inclusion bodies. The fully functional peptibodies require the form of 6 disulfide bonds including two interchain disulfide bonds between two monomeric units and 4 intra-chain disulfide bonds and need to be converted into homodimers during the refolding process. The inclusion bodies were isolated from *E. coli* cells and denatured in the presence high concentration of denaturing agents such as GdHCl and Urea. The denatured peptibodies were continuously diluted in refolding buffer using a tubular reactor, allowing for a defined refolding time for conversion of monomer to homodimer. The outlet of the tubular reactor was directly connected to the protein A affinity chromatographic column, where refolded peptibodies were directly captured, washed, and eluted as a soluble and refolded form. To optimize the refolding process parameter and get the best operating condition, the central composite design was performed. The purity and the refolding yield of the eluted sample were calculated using different orthogonal techniques. The refolding yield was found more than 50% using the optimized oxidative refolding conditions and different residence times. Not only purity was more than 80%, but it was also correctly folded too. The 3-dimensional structure and formation of correct disulfide bond were measured using Fluorescence and High dimension Liquid chromatography-mass spectrometry (LCMS) respectively.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Rupali Kumthekar
(CSIR-NCL)

Session Category: Biophysical and Analytical Characterization Technologies II

Title: Mapping of disulfide linkages using CID based mass spectroscopic approach for peptibody: A Fc-fusion proteins

Abstract: Disulfide bonds are commonly found in covalent interactions that play a vital role in establishing the structure of proteins and maintaining their biological activity. Thus, the characterization of disulfide linkages is essential to ensure the safety and effectiveness of biotherapeutic drugs. This investigation is focused on mapping intra and inter-disulfide bond formation during the in-vitro refolding of recombinant peptibody using LC-ESI-MS/MS. The selected recombinant peptibody is a homodimeric, a-glycosylated Fc-fusion protein expressed in a bacterial host system. Monomers refold to form CH2 and CH3 scaffold, each domain consisting of one intra-chain disulfide bond. The two monomers had connected through two inter-chain disulfide linkage. The intact mass was determined for the biosimilar with a mass error of <1Da compared to the expected mass. However, subunit mass was determined under both reduced and reduced alkylated conditions using LC-ESI-MS. The amino acid sequence in the disulfide bond containing peptides was confirmed at MS/MS levels using the CID- based approach. Furthermore, a bottom-up mass spectroscopy approach was used for mapping inter-chain (Cys7-Cys7and Cys10-Cys10) and intra-chain (Cys42-Cys102 and Cys148-Cys206) disulfide bonds of therapeutic peptibody. The secondary and higher order structure was found to be indistinguishable from innovator. The combination of orthogonal analytical approaches using intact mass analysis, sub-unit mass analysis and peptide fingerprinting using CID-based fragmentation helps in rapid and accurate confirmation of disulfide bond position in biosimilar and innovator molecules. Observations from this study will help characterize structural integrity and quality during the process and product development for consistent product quality.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Nandita Srivastava
(IMTECH, Chandigarh)

Session Choice: Bioprocessing for Nutraceuticals

Title: Polysaccharide-based self-assembled smart hydrogel for in vitro delivery of co-encapsulated probiotics and folic acid

Abstract: The biologically active platforms such as hydrogels are three-dimensional hydrophilic porous structures capable of supporting bioactive components attachment to their surface. They can serve as cargos for sustainable and controlled delivery of compounds to the site of action through reversible sol-gel transitions. Hence, it is vital to develop novel hydrogel materials with smart or intelligent properties that can undergo structural and volume changes by responding to external triggers. Therefore, we have fabricated a self-assembled, stimuli-responsive, novel tri-composite polysaccharide-based hydrogel of chitosan, gellan, and κ-carrageenan devoid of toxic chemical cross-linkers. A polysaccharide-based composite hydrogel can overcome problems associated with individual polysaccharide gels, such as mechanical strength and stability. Although, there are several reports on encapsulation of probiotics and bioactive compounds, however maintaining the viability of loaded probiotics, stability of folate, and their bioavailability is still a challenge. We utilized tri-composite hydrogel to facilitate co-encapsulation of folate (vitamin B9) and probiotic spores. Its co-encapsulation with folate, while increasing nutritional value of the system, also helps to maintain viability of spores. The studies on hydrogel revealed that it had remarkable encapsulation efficiency and exhibited sustainable release. The release rate studies at different pH suggested maximum release in alkaline condition, which correlates with release in vitro in the simulated intestinal phase. Moreover, microscopic and FE-SEM analysis depicted conversion and colonization of bacterial spores to vegetative cells in the intestinal phase. Overall, this study paves the way to develop a single green matrix for co-encapsulating various functional foods with controlled delivery at the targeted site.

Day 3, Sunday, December 18, 2022

Session 20

Dr. Kanti Mihooliya,
IIT Delhi

Session Choice: Protein Unfolding, Refolding and Aggregation

Title: Optimization of process intermediates conditions to enhance stability and tackling hold-ups during purification: L-Asparaginase as a case study

Abstract : L-Asparaginase is a vital enzyme in the food and biopharmaceutical industries. It catalyzes the hydrolysis of L-asparagine amino acid into aspartic acid and ammonia, which became the basis of its exploitation in treating acute lymphoblastic leukemia (ALL) and acrylamide mitigation from baked and fried food products. The commercially available asparaginase is catalytically active in its tetrameric form, which is considered dissociated during different manufacturing and storage conditions. Hence, the stability of L-asparaginase is a known issue, making it an expensive molecule. Downstream processing of any biopharmaceutical usually involves hold-ups during various purification steps because of manual or instrumental error and design limitations, resulting in loss of the final yield of the product. Therefore, the stability of L-asparaginase must be enhanced to tackle the hold-up times and boost the final yield. In the present study, we propose a strategy to increase the L-asparaginase yield and stability to tackle the hold-ups during different stages of its downstream processing. An OVAT followed by a design of experiment-based optimization of solubilization and refolding process resulted in a 3-fold increase in enzyme recovery compared to unoptimized conditions. L-asparaginase stability under the optimized conditions showed that the refolding output was stable for more than a week, while the control sample showed no activity after 72 h. The current study highlights that for products that are liable to degradation, product stability needs to be given due consideration along with the traditional criteria of step recovery and other product quality attributes.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Rashmi Sharma,
IIT Delhi

Session Choice: Protein Unfolding, Refolding and Aggregation

Title: Optimization of the in-vitro refolding of biotherapeutic Fab Ranibizumab

Abstract: Recombinant biotherapeutics expressed as inclusion bodies require solubilization and subsequent refolding to attain a functionally active state. But the refolding step often acts as a bottleneck due to a low throughput, cumbersome and expensive framework, especially in the case of complex, multi-domain proteins such as antibody fragment (Fab) molecules. Fab Ranibizumab, with two domains and five disulphide bonds, currently presents a daunting task to be refold. In this paper, the in-vitro refolding of this Fab over 24 hours was investigated, using two different methodologies. The first protocol employed the traditional two-step DoE for screening and optimization. The entire refolding process was considered as a single process step. The second approach derived inferences from the data of analytical tools like intrinsic fluorescence, zeta potential and RP-HPLC to highlight a possible time-based molecular behavior during refolding. This led to the identification of a breakpoint at the 8th hour of the process, proposing initial occurrences of the native tertiary conformation. Based on this observation, segmented DoEs were conducted to optimize the two time zones of the refolding process (0-8 hours and 8-24 hours). This unconventional segment-based optimization approach led to a 55% increment over the standard conventional optimization methodology, performed for the same Fab, Ranibizumab. It prevailed with an effective yield of 32% over the conventional strategy with 20.6%.

Day 3, Sunday, December 18, 2022

Session 20

Mr. Santosh G. Ughade,
CSIR-NCL, Pune

Session Category: Protein Unfolding, Refolding, and Aggregation

Title: Cloning, Expression, and Refolding of Recombinant Therapeutic Peptides

Abstract: Cloning is the best platform to produce human-animal protein in the bacterial host system by using recombinant DNA technology. *E. coli* is the most frequently used bacterial host system for the production of therapeutic active protein. In industries, with large protein production at low cost and less time *E. coli* is preferable for its genetic simplicity, expensive, and fast high-density cultivation. In bacterial host systems lack of post-translation modification, and disulfide bond formation may lead to inclusion bodies formation. We are focused on the large quantity of protein production by overexpressing the protein gene in form of inclusion bodies. Inclusion bodies are the aggregated form of protein and they need to further refold protein by giving a suitable environment for protein refolding after the solubilization of inclusion bodies by using chaotropic agents. Production of a small therapeutic peptide like insulin, teriparatide, etc. This investigation is focused on the understanding effect of expression construct, codon optimization, and induction strategies for protein production at different pH and temperatures. Successfully recovering insulin lispro from inclusion bodies is the crucial and key step to guarantee high efficiency for the manufacturer. To achieve a high yield of protein folding we have developed the refolding buffer and refolding parameters for specific peptides. There are two by-products of disulfide-linked oligomer and disulfide isomerized monomer that are identified by using various analytical techniques.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Anupa,
IIT Delhi

Session Choice: Downstream Processing for Recombinant Protein Therapeutics

Title: Enablers of continuous processing for biotherapeutic products

Abstract : The benefits of continuous processing over batch manufacturing are widely acknowledged across the biopharmaceutical industry, primary of which are higher productivity and greater consistency in product quality. Further, the reduced equipment and facility footprint lead to significantly lower capital costs. Technology enablers play a major role in this migration from batch to continuous processing. Here, various enablers that are facilitating adoption of continuous upstream and downstream bioprocessing are highlighted. This includes new bioreactors and cell retention devices for upstream operations, on column and continuous flow refolding, and novel continuous chromatography, and single pass filtration systems for downstream processes. The significant roles of process integration and control as well as of data analytics have also been briefly discussed.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Anuja Prabhu,
CSIR-NCL, Pune

Session Choice: Upstream Processing for Recombinant Protein Therapeutics

Title: Engineering nucleotide sugar synthesis pathways for independent and simultaneous modulation of N-glycan galactosylation and fucosylation in CHO cells

Abstract : In therapeutic recombinant glycoproteins like monoclonal antibodies (mAbs), glycosylation is a critical quality attribute. Glycosylation can affect effector functions of these glycoproteins and therefore, their therapeutic use can depend on a particular glycoform profile. In this study, we demonstrate the ability to predictably modulate glycosylation in recombinant protein, which can aid in achieving a targeted glycoform profile and thereby desired clinical efficacy. We have engineered nucleotide sugar synthesis pathways in CHO cells using CRISPR/Cas9 system for combinatorial modulation of galactosylation and fucosylation levels by simply varying extracellular galactose and fucose concentration. UDP-Gal and GDP-Fuc are the nucleotide sugar donor substrates for galactosylation and fucosylation. These can be synthesized either via de novo pathway from glucose or via salvage pathway from sugars. In CHO cells expressing recombinant IgG, UDP-galactose 4′-epimerase (Gale) and GDP-L-fucose synthase (Fx) enzymes were knocked out to block the de novo synthesis of UDP-Gal and GDP-Fuc. This makes their synthesis dependent on salvage pathway, which relies on extracellular availability of galactose and fucose. In Gale knock-out (KO), the array of N-glycans seen in the parental host were restricted to predominantly agalactosylated A2F glycan (89%), whereas in the Gale/Fx double KO to agalactosylated and afucosylated A2 glycan (88%). With galactose and fucose supplementation to Gale/Fx KO, the glycoform profile could be modulated from comprising predominantly agalactosylated and afucosylated glycans to up to 42% galactosylation and 96% fucosylation in a sugar concentration dependent manner. Thus, using a single host system, we show that galactosylation & fucosylation levels can be simultaneously and independently varied across a wide range by simply varying extracellular supplementation of sugars. Such an engineered CHO host platform can not only help in achieving predefined glycoform, but can also be useful for rapid synthesis of variably glyco-engineered proteins for early examination of optimal glycoform for desired biological activity.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Rucha Patil,
IIT Delhi

Session Choice: Upstream Processing for Recombinant Protein Therapeutics

Title: Optimization of process parameters for enhanced production of ranibizumab in *E. coli*.

Abstract : Microbial, specifically *E. coli*-based systems, are favoured hosts for recombinant protein expression. However, low expression levels of fab fragments, and the resulting high manufacturing cost, is one of the significant challenges that the manufacturers face. For such cases, development of a robust, well characterized upstream process becomes a necessary requirement if one is to achieve favourable process economics. This paper presents a systematic approach for optimization of a *E. coli* based fermentation process for production of a fab fragment, ranibizumab. A two-step approach has been used. Six variables were examined: post-induction temperature, post-induction time, inducer concentration, agitation speed, media pH, and induction OD. These parameters were evaluated for their effect on biomass concentration, protein titre, impurity level, and light to heavy chain ratio. First, a screening DOE with fractional factorial design has been used to shortlist important process parameters. Next, this has been followed by a response surface study based on central composite design for identifying the operating range for the significant process parameters. Induction OD, IPTG concentration, media pH and temperature were identified as significant parameters with their respective optima at 1.25 OD, 1.375 mM, 7 pH, and 28.5°C, respectively. Operating under these conditions resulted in formation of 0.046 g protein/g IB, 5.2 g/L biomass concentration and equal expression ratio (LC:HC=1.1), with 8% impurity. Additionally, based on the predictions, we found that temperature, induction OD, and their interactions significantly affect the LC:HC ratio and impurity level, and need to be tightly controlled. Temperatures over 25°C with early induction up to mid-log phase are preferred.

Day 3, Sunday, December 18, 2022

Session 20

Mr. Sami Ullah Bhat,
IIT Delhi

Session Choice: Upstream Processing for Recombinant Protein Therapeutics

Title: A real time feeding control strategy for enhanced recombinant protein production and acetate control in *E. coli* using near infrared spectroscopy (NIRS) as a PAT tool

Abstract: Near-infrared spectroscopy (NIRS) has been used for real-time quantification of lactose, acetate, and galactose in fed-batch cultivation of *E. coli* with glycerol and lactose feeding. This has been used as part of a process analytical technology (PAT) control system for real-time control of acetate, a by-product of *E. coli* overflow metabolism and a critical process parameter known to inhibit cell growth and recombinant protein production. Acetate concentration was controlled in real-time by automatically reducing the glycerol feed rate whenever acetate concentrations were observed to exceed certain fixed thresholds. The control system ensured that acetate did not accumulate above 10 g/L, a limit found to be effective for enhancing recombinant protein production in the current process. Lactose feeding was also controlled in real-time with minimal accumulation by triggering pulse feeding as soon as the concentration in the bioreactor dipped below a threshold of 2 g/L. Production of recombinant protein rPramlintide was enhanced by 64% in the PAT-controlled process. The proposed control system is robust and well suited for applications where under-feeding of glycerol is conventionally used to prevent acetate accumulation at the expense of a potential increase in product yield.

Day 3, Sunday, December 18, 2022

Session 20

Ms. Priya Mishra,
IIT Bombay

Session Choice: Upstream Processing for Recombinant Protein Therapeutics

Title: Development of a recombinant cell line with increased secretory capacity by Tunicamycin Adaptation

Abstract : Biotherapeutics or biologics are complex active molecules obtained from biological sources used for the diagnosis, prevention, treatment and cure of diseases. Mammalian cell lines such as CHO are widely used for the production of biotherapeutic molecules. Various engineering approaches are used to improve the characteristics of host cells to achieve a high titre and productivity of the desired product. Our lab has earlier reported that the induction of ER stress by tunicamycin (TM) adaptation activates the UPR pathway by upregulating the expression of various chaperones and transcription factors. This led to an increase in IgG productivity by two folds in recombinant CHO cells. The universality of the effect of TM adaptation was tested on an in-house recombinant CHO-K1 cell line, RBD-E1, producing the RBD domain of the Spike protein (SARS-CoV2), with an aim to increase the productivity. A high-density culture was achieved by adapting the cells to SFM suspension culture. Further RBD-E1 cells were adapted to increasing conc. of TM, and over the subsequent round of adaptation, the viability drop post TM treatment was decreased. To understand the mechanistic basis of increased TM resistance, the expression level of chaperones and transcription factors of the UPR pathway was measured. No significant increase in the mRNA levels was seen; however, a 40-fold upregulation in the TM target gene – DPAGT1, which is the primary enzyme of the glycosylation pathway, was observed. Similar results were observed when cells were adapted to TM using an alternate adaptation method. This suggests that there are potential multiple modes of adaptation to TM. The effect of TM adaptation on protein productivity is under investigation.

Day 3, Sunday, December 18, 2022

Session 20

Mr. Pavan Allampalli,
IIT Guwahati

Session Choice: Upstream Processing for Recombinant Protein Therapeutics

Title: Metabolic heat rate based soft sensors for real-time estimation of specific growth rates and implication of process control strategies for the production of human interferon alpha 2b

Abstract : Implementation of Process Analytical Technology (PAT) in the biopharma industry is severely plagued due to the non-availability of reliable online sensors which monitor the critical process parameters (CPPs). The present investigation was focused on the development of a soft sensor for the real-time estimation of specific growth rates. The metabolic heat rate deciphered from the fermentation calorimeter was used to model two different specific growth estimators. The basis for the first estimator was the dissipation of instantaneous microbial heat rate. The second estimator was formulated by looping the cumulative metabolic heat and biomass heat yield coefficient into the estimator model. The raw data emanated from the calorimetric signal was processed with digital filters to minimize the noise. Estimators based on cumulative heat resulted in better performance with lower RMSE values (0.031 – 0.046) for yeast (Pichia pastoris) and bacterial systems (Streptococcus zooepdemicus). Further, this estimator was coupled with three different control strategies envisaged to control the specific growth rate of glycoengineered P. pastoris to produce interferon α2b. A pre-determined feeding of limiting substrate (methanol) was performed through a feedforward control strategy at desired setpoint values of 0.04, 0.035, and 0.03h-1, respectively. The second control strategy was enabled with a feedback loop in the form of standard PID control. Finally, an adaptive PID control configured gain scheduler was incorporated to maintain tight control over the optimal specific growth rate (0.035h-1). Robust control of methanol feeding invoked by adaptive PID yielded a 1.5- and 2.2-fold increase in productivity of interferon α2b compared to the other two control strategies.

Day 3, Sunday, December 18, 2022

Session 20

Dr. Sambit Sarkar

Session Category: Upstream Processing for Recombinant Protein Therapeutics

Title: Downstream processing of chlorophylls, proteins and carbohydrate from microalgae biomass using three phase partitioning.

Abstract : Microalgae are promising feedstock for different valuable biomolecules such as carbohydrate, proteins and chlorophylls. Chlorophylls in various forms are marketed as nutraceuticals due to its positive effects on human health. Chlorophylls are also marketed as food coloring agents such as E140 and E141. On the other hand, microalgae proteins are considered as alternative proteins in different food and food supplements. Proteins or carbohydrates are not considered as standalone products for large scale microalgae cultivation due to comparative low market value of proteins and carbohydrates and relatively higher production cost of microalgae. The production of microalgae becomes economically feasible option if carbohydrates and proteins are considered as the byproduct along with the high valued chlorophylls as main product. In this work, a modified version of three phase partitioning (TPP) was investigated for the simultaneous fractionation of chlorophylls, carbohydrates and proteins from the biomass of isolated microalgae. Recovery yield of 71%, 40% and 22% was recorded for proteins, carbohydrates and chlorophylls respectively, with optimised homogenisation assisted tri-phasic separation. A comparative analysis between homogenisation assisted-TPP and TPP without homogenisation was carried out. Significant improvements were observed in terms of recovery yields in the case of HTPP compared to TPP (38% for proteins, 14% for carbohydrates and 10% for chlorophylls).

Day 3, Sunday, December 18, 2022

Session 20

Mr. Ashish Kumar

Session Category: Bioprocessing for Natural Medicinal Products

Title: CRISPR-cas9 mediated generation of lanosterol deficient yeast strain for the efficient production of triterpene-related products

Abstract: Euphorbia plant species have very prominent and diverse triterpenes. Euphol and tirucallol are the two significant and majorly present tritetriterpene in E. tirucalli and E. grantii. These two triterpenes have anti-cancerous, anti-bacterial, and antifungal biological activities. The functional characterization of genes responsible for the biosynthesis of euphol and tirucallol has been done but has yet to be with the proper standards and how they have identified this gene. To determine the two triterpene cyclases, transcriptome analysis of the leaf and stem tissue of E. tirucalli and E. grantii has been performed. This led us to identify thirteen triterpene cyclases (TTS). Based on NCBI blastx analysis, eight were full-length out of thirteen triterpene cyclases. Sequences Phylogenetic and active site amino acid analysis identified two identified β-amyrin synthases (EugTTS6, EugTTS7) and that were proved to be cycloartenol synthase (EugTTS8). Five unique and unidentified full-length TTS were cloned and functionally characterized in YPH499 and lanosterol deficient yeast (TMBL17) expression system. These five triterpenes are were described as α-amyrin synthase (EutTTS1), lupeol synthase (EutTTS2), friedelin synthase (EutTTS3), euphol tirucallol synthase (EutTTS4), and euphol synthase (EutTTS5).

Day 3, Sunday, December 18, 2022

Session 20

Dr. Balaji Sitharaman

Session Category: Advance Biomanufacturing capabilities: Next Generation Technology Solutions

Title: All Carbon Stationary Phase Material for Biologic Separation: Design and Characterization

Abstract: Synthetic graphitic carbon and bonded silica stationary phase materials are the earliest studied media for reverse-phase liquid chromatography. This presentation highlights recent developments in carbon-based stationary phase media using natural graphite as the starting material. First, an overview and unique capabilities of this material's synthesis platform will be depicted. Next, a summary of the fundamental physicochemical properties of the natural graphitic carbon-based stationary phase media will be presented. Further, its differentiation over the current state-of-art reverse-phase chromatography media will be elucidated. Finally, using insulin, semaglutide, bovine serum albumin, and hemoglobin as examples, chromatography performance studies will be presented.

Day 3, Sunday, December 18, 2022

Session 21

Title: Novel Gene Therapy for Cancer cure: an Experience from Laboratory to Clinic

Dr. Rahul Purwar

Abstract

Chimeric Antigen Receptor-T cells (CAR-T cells), a type of gene therapy, has shown remarkable success in curing hematological cancers. However, complexity of the CAR-T cells manufacturing and high costs (4-5 crores INR per patient) of the available therapy limit their accessibility in low resource settings including India. Here we will discuss our; a collaborative effort of IIT Bombay and Tata Memorial centre (TMC) Mumbai, experience of design, development and clinical validation (Phase I trials, first in-human) of indigenously developed CAR-T cell therapy in India. In brief, a novel HCAR19 product as well patient-scale manufacturing process were designed and developed in an integrated functionally closed system at cGMP facility of IIT-Bombay and recently at ImmunoACT, a spin off company of IIT Bombay. To examine the feasibility and functionality of the developed manufacturing process and platform, the end-to-end CAR-T cell manufacturing (including the lentiviral vector and QC) was performed for two Phase I clinical trials. HCAR19 cells from all 17 patients were manufactured successfully within 8 days of culture (transduction efficiency: >25%, dosage range: 1.78 x 106 - 17 x 106 viable HCAR19/kg). The final HCAR19 product showed robust expansion (20 - 30 folds) and maintained the presence of anti- tumor cell subsets like naïve and central memory T cells. Final product from all 17 patients qualified the release criteria for safety, identity, purity and potency as perdefined specification. The unit economics in fully-scaled production model was approximately 30,000 USD (or 25,00,000 INR) per dosage for the end-to-end CAR-T cell delivery including logistics, lentiviral vector cost, manufacturing costs and quality control. In conclusion, we developed a novel anti-CD19 CAR-T cell product and developed highly affordable CAR-T cell therapy platform. HCAR19 is safe and active with manageable toxicities in patients with refractory large B-cell lymphoma. The favorable safety profile may allow delivery in an outpatient setting and in a cost-effective manner particularly critical to the delivery of CAR T-cells in LMIC countries.Given the benefit of affordable indigenous platform to broader socio-economic baseof patients, the cell and gene therapy will be poised for a transformational change as a mass therapy.

Day 3, Sunday, December 18, 2022

Session 21

Title: Industrializing the Cell and Gene Therapy Ecosystem.

Dr. Likhesh Sharma

Abstract

Cell therapies are one of most complex therapies as they often require cellular engineering therefore the manufacturing processes have high cost and pose unique challenges for the industry. In this talk I would give an overview of current industrial landscape of cell therapies, challenges and few case studies on how these challenges were addressed by Cytiva and how digital innovation and further pave the path ahead.

Day 3, Sunday, December 18, 2022

Session 21

Title: The role of biomaterials in stem cell therapeutics and biomanufacturing

Dr. Ganesh ingawale

Associate professor/dbt-ramalingaswami fellow
Symbiosis center for stem cell research (scscr),
Symbiosis school of biological sciences (ssbs),
Symbiosis international (deemed university), lavale, pune - 412115

Abstract:

Tissue engineered biomaterial scaffolds or bioreactors have enormous promise for cell culturing for future target biotherapeutics. Tissue abnormalities and disorders have become more common in recent years. The advancement of regenerative medicine focuses attention on stem cell-biomaterial-based therapies. Tissue engineering and regenerative medicine offer the strategy of producing spatially, physico-chemically, and physiologically tailored biomaterials for stem cell growth and differentiation. This talk will provide a brief overview of several strategies of combining stem cells and biomaterials for tissue engineering applications in terms of bone, cartilage, and skin tissue regeneration. Further, this talk would also discuss different stem-cell-related biomanufacturing approaches and the novel bio-fabrication technologies including stem-cell secretome (extracellular vesicles), enlightening a promising route for the future advancement of large-scale stem cell-biomaterial-based therapeutic manufacturing.