



The 10th RPP Conference was held in Crete, Greece from 24 to 27 April. It addressed a topic with major implications for on modern society, as recombinant proteins are used in a wide range of vital processes, including the processing of food and beverages, the development of important medicines, and the sustainable production of chemicals and biofuels. As a result, there is a constant need for new production platforms that can deliver protein products at higher yields, better quality and at lower cost. The RPP conferences address these challenges by offering comprehensive overviews of current bottlenecks and innovative solutions in recombinant protein production across different cell factories. Special attention is attributed to the comparative physiology of microbial and higher eukaryotic protein production platforms. The RPP conferences were built on the successes of previous meetings, especially the many companies that support these meetings.

The topics included:

- Host physiology under recombinant protein production conditions
- Cell engineering for recombinant protein production
- Recombinant protein production processes at industrial scale
- New format and difficult to express proteins

# EXTENDED PROGRAMME

## Wednesday 24th of April 2019

09:00                      *Registration opens (Creta Maris, Crete)*

### Special PreConference Session (ProteinFactory, BioToP)

- 10:30 - 10:40              Welcome address and introduction of the ProteinFactory and BioTop PhD programs.
- 10:40 - 10:55              **Daphne Mermans** (University of Kent, United Kingdom, Stockholm University, Sweden)  
  
*Co-translational folding of proteins translocated by the *Escherichia coli* Twin Arginine pathway monitored *in vivo* and *in vitro*.*
- 10:55 - 11:10              **Margarita Bernal Cabas** (University of Groningen, the Netherlands)  
  
*In vitro* characterization of TatAyCy protein translocase complexes from *Bacillus subtilis*.
- 11:10 - 11:25              **Minia Antelo** (University of Greifswald, Germany)  
  
Absolute quantification of genome reduced *Bacillus subtilis* under secretion stress: a mass spectrometry-based proteomics approach.
- 11:25 - 11:40              **Ana Bulovic** (Humboldt-Universität zu Berlin, Germany)  
  
A whole-cell model of *E. coli* as an *in silico* experimental tool for recombinant protein production.
- 11:40 - 11:55              **Sonakshi De** (BOKU, Austria)  
  
Low growth rate triggered transition to pseudo-hyphal phenotype of the protein production host *Pichia pastoris*.
- 11:55 - 12:10              **Dominik Jeschek** (BOKU, Austria)  
  
A toolbox to express and characterize membrane transport proteins

## Wednesday 24th of April 2019

### Special PreConference Session (ProteinFactory, BioToP)

- 12:10 - 12:25      **Philipp Mundspurger** (BOKU, Austria)  
HIV-1 envelope glycoproteins as vaccine antigens - a critical reflection on expression and manufacture.
- 12:25 - 12:40      **Nina Bydlinski** (BOKU, Austria)  
Enhancement by reduction: pushing N-glycosylation in CHO cells.
- 12:40 - 14:00      *Lunch*

### Welcome and opening session

- 14:00 - 14:20      *Organisers' opening and welcome address*  
**Diethard Mattanovich** (BOKU, Austria)  
**Colin Robinson** (University of Kent, United Kingdom)
- 14:20 - 14:50      *Opening keynote lecture*  
**Nathan Lewis** (University of California, San Diego, USA)  
Unraveling the protein-specific requirements in mammalian protein expression.

### Session 1: Host physiology under recombinant protein production conditions

Chair: Diethard Mattanovich

- 14:50 - 15:20      **E. Tjeerd van Rij** (DSM, the Netherlands)  
Exploring synthetic expression modules with FACSseq in *Bacillus subtilis*.
- 15:20 - 15:40      **Tobias Klein** (BASF, Germany)  
The secretion stress response genes *htrA* and *htrB* are modulated by the choice of signal peptide during protein secretion in *Bacillus*.
- 15:40 - 16:00      **Joseph Shiloach** (National Institutes for Health, USA)  
Effect of over expressing protective antigen on global gene transcription in *Bacillus anthracis* BH500.
- 16:00 - 16:30      *Coffee break*

## Wednesday 24th of April 2019

### Session 1: Host physiology under recombinant protein production condition

Chair: Diethard Mattanovich

- 16:30 - 17:00      **Francesca Ceroni** (Imperial College, United Kingdom)  
The burden of recombinant protein production: a synthetic biology perspective.
- 17:00 - 17:20      **Rosa Morra** (University of Manchester, United Kingdom)  
Excretion of cytoplasmic proteins: are you missing out?
- 17:20 - 17:40      **Ken-Ichi Yoshida** (Kobe University, Japan)  
*myo*-Inositol-1-phosphate synthase restored in *Bacillus subtilis* to produce *scyllo-inositol*, a therapeutic agent for Alzheimer's disease, from glucose.
- 17:40 - 18:00      **Fabien Letisse** (LISBP Toulouse, France)  
Dynamic metabolic response to heterologous protein overproduction in *E. coli*.
- 18:00 - 18:20      **Ross Dalbey** (Ohio State University, USA)  
The mechanism of membrane insertion by the bacterial YidC.
- 18:20 - 21:00      *Poster setup, exhibition and welcome buffet*

## Thursday 25th of April 2019

### Session 2: Cell engineering for recombinant protein production

Chair: Colin Robinson

- 09:00 - 09:30      **Kerstin Otte** (Biberach University of Applied Sciences, Germany)  
Engineering of mammalian cell lines to remove production bottlenecks.
- 09:30 - 09:50      **Lise Marie Grav** (Novo Nordisk Foundation Center for Biosustainability, Denmark)  
Minimizing clonal variation during mammalian cell line engineering for improved systems biology data generation.
- 09:50 - 10:10      **Ebru Alazi** (Dutch DNA Biotech BV, the Netherlands)  
Rational design of fungal strains for production of plant biomass degrading enzymes.
- 10:10 - 10:30      **Andreas Knapp** (Heinrich Heine University Düsseldorf, Germany)  
Small changes with big impact - how single nucleotide or amino acid substitutions affect production and secretion of recombinant proteins by *Bacillus subtilis*.
- 10:30 - 11:00      *Coffee break*
- 11:00 - 11:30      **Patrick Stargardt** (enGenes Biotech, Germany)  
Growth-decoupled recombinant protein production in *Escherichia coli*.
- 11:30 - 11:50      **Douglas Browning** (University of Birmingham, United Kingdom)  
The rise of garage science: making recombinant protein production as cheap as chips.
- 11:50 - 12:10      **Julie Zedler** (University of Copenhagen, Denmark)  
A fast-growing cyanobacterium: new hope for green cell factories.

## Thursday 25th of April 2019

### Session 2: Cell engineering for recombinant protein production

Chair: Colin Robinson

- 12:10 - 12:30      **Rocio Aguilar Suárez** (University Medical Center Groningen, the Netherlands)  
Going smaller: how “debugged” *Bacillus subtilis* behaves as a cell factory.
- 12:30 - 13:30      *Lunch*
- 13:30 - 15:00      *Poster Session: odd posters*

### Session 2: Cell engineering for recombinant protein production

Chair: Jan Maarten Van Dijl

- 15:00 - 15:20      **Markku Saloheimo** (VTT Technical Research Center, Finland)  
Development of the filamentous fungus *Myceliophthora thermophila* C1 into a next-generation therapeutic protein production system.
- 15:20 - 15:40      **Jasmin Fischer** (Bisy, Austria)  
Inactivation of cell wall proteins to enhance protein secretion.
- 15:40 - 16:00      **Alexander Frey** (Aalto University, Finland)  
Nature driven design of yeast-based cell factories.
- 16:00 - 16:30      *Coffee break*
- 16:30 - 16:50      **Andreas Licht** (Jena Biotech GmbH, Germany)  
Humanizing the glycosylation pattern of the *Leishmania* expression system LEXSY.
- 16:50 17:10      **Lasse Pedersen** (Technical University, Denmark)  
CRISPR/Cas9 library screen >16000 gRNAs against CHO.
- 17:10 – 17:30      **Gero Steinberg** (University of Exeter, UK)  
A cell biology approach towards understanding secretion in *Trichoderma reesei*.
- 17:30 – 18:10      **Nicole Borth** (BOKU, Austria)  
Darwin and Lamarck unified: how to control phenotypes without changing the genome sequence.

## Friday 26th of April 2019

### Session 3: Recombinant protein production processes at industrial scale

Chair: Pau Ferrer

- 09:00 - 09:30      **Andy Racher** (Lonza, United Kingdom)  
Shorter timelines for DNA to the first clinical batch: the importance of good process development.
- 09:30 - 09:50      **Johanna Jarmer** (Boehringer Ingelheim RCV, Austria)  
Enhanced soluble expression in *Escherichia coli* and miniaturized fed batch fermentation screening platform.
- 09:50 - 10:10      **Núria Adelantado** (Lonza Pharma and Biotech, UK)  
*XS™ Pichia 2.0*: closing the gap between bacterial and mammalian systems for the production of recombinant proteins.
- 10:10 - 10:30      **Peter Neubauer** (Technical University Berlin, Germany)  
Model based development of bioprocesses with recombinant microorganisms: adaptive model-based designs for efficient parameter estimation in parallel bioreactors.
- 10:30 - 11:00      *Coffee break*
- 11:00 - 11:20      **Allan Kent Nielsen** (Novozymes, Denmark)  
Heterologous expression of amylase and PrsA pairs and their influence on the secretion stress response in *Bacillus subtilis*.
- 11:20 - 11:40      **Roland Weis** (Validogen, Austria)  
Productivity through diversity - a protein production toolbox to UNLOCK *Pichia*.
- 11:40 - 12:00      **David Wollborn** (RWTH Aachen, Germany)  
Small scale cultivation strategy ensuring the optimal induction time point for individual *Komagataella phaffi* clones during primary screening.
- 12:00 - 13:00      *Lunch*
- 13:00 - 15:00      *Poster Session: even posters*

## Friday 26th of April 2019

### Session 4: New format and difficult to express proteins

Chair: Jeff Cole

- 15:00 - 15:30      **Emma Davé** (UCB Celltech, United Kingdom)  
Considerations for efficient production of Fab-based bispecific antibodies.
- 15:30 - 15:50      **Johanna Hausjell** (Technical University Viena, Austria)  
Challenge accepted: recombinant production of hard-to-express membrane-bound cytochrome P450 in yeast.
- 15:50 - 16:10      **Myrsini Michou** (National Hellenic Research Foundation, Greece)  
SuptoXD/SuptoxR: two specialized *Escherichia coli* strains for high-level recombinant membrane protein production.
- 16:10 - 16:40      *Coffee break*
- 16:30 - 16:50      **Lloyd Ruddock** (University of Oulu, Finland)  
Sweet applications of disulfide bond formation in the cytoplasm.
- 17:00 17:20      **Rochelle Aw** (Imperial College London, United Kingdom)  
Engineering a novel *Pichia pastoris* cell free protein synthesis platform.
- 17:10 – 17:50      **Guido Grandi** (University of Trento, Italy)  
Synthetic biology of bacterial outer membrane vesicles (OMVs) for the production of vaccines against infectious diseases and cancer.
- 17:50              *Closing of the scientific programme. Concluding remarks*
- 20:00              *Gala Dinner*