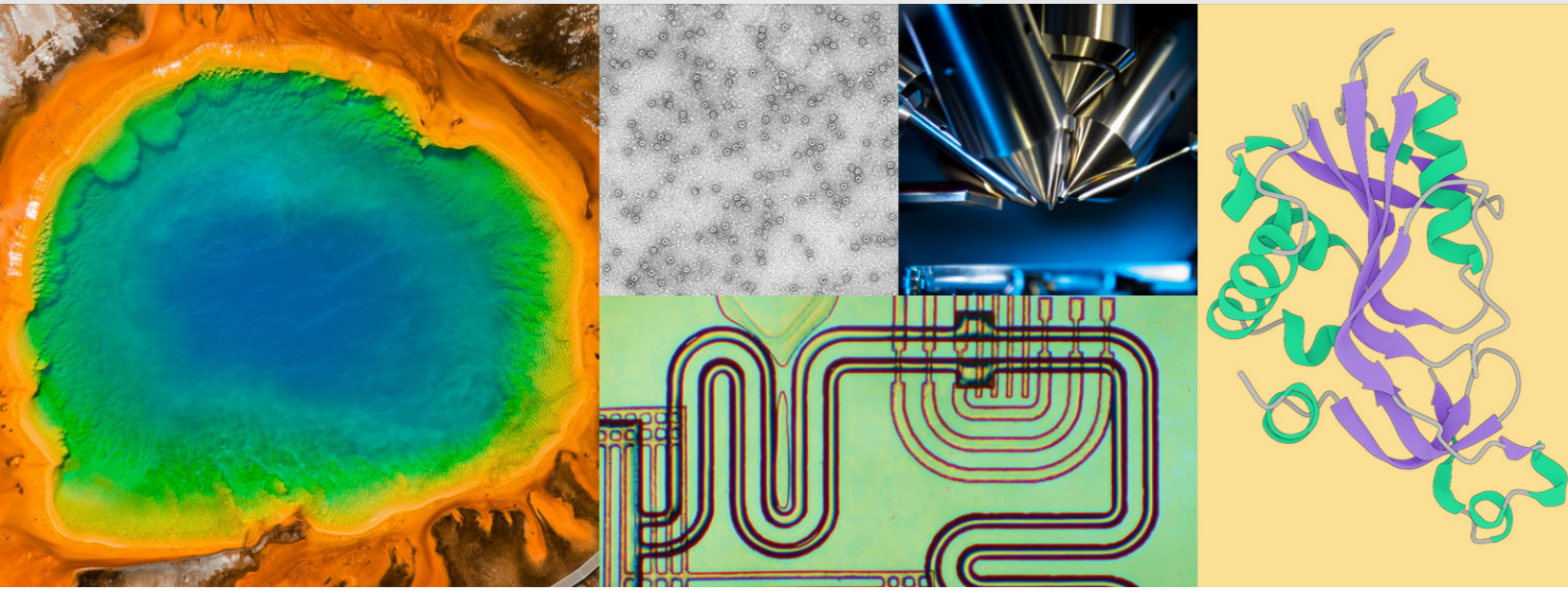
PROHITS

Prokaryote proteomics at high temperature for single cells



Issue 1
July 2025

This issue of the newsletter has been edited by Josephine Boel Andresen (DC2) and Dinu Zinovie Ciobanu (DC6).

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PROHITS at a glance

The project in a nutshell

PROHITS is a Marie Skłodowska-Curie Doctoral Network (MSCA-DN) project with the overall aim of resolving the proteome of prokaryotes at single-cell level and at a range of temperatures. This will be achieved by integrating experimental and computational approaches, and will improve our understanding of thermophile biology to optimally design cell factories and the related industrial processes.

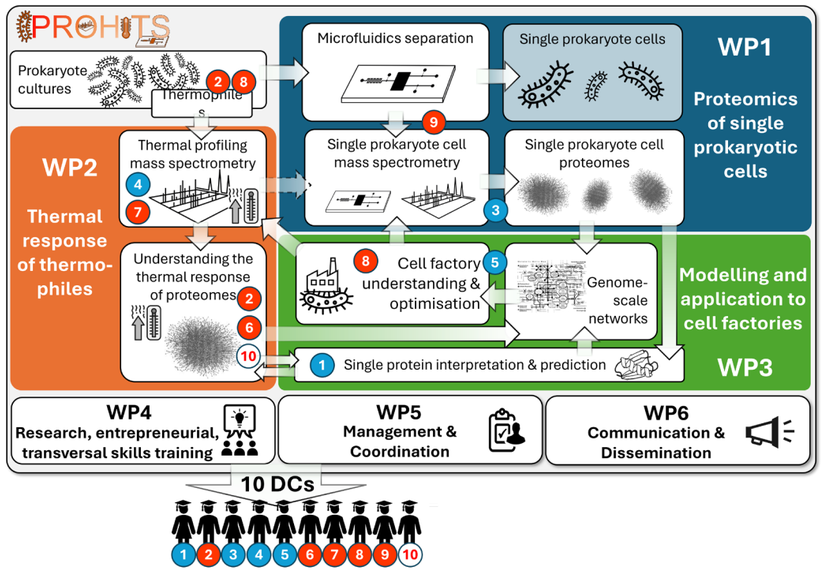
The PROHITS DN will advance 1) microfluidics to separate single prokaryotic cells, 2) mass spectrometry (MS) and its associated bioinformatics to handle single prokaryotic cells, and 3) MS-based thermal proteome profiling to study the thermal response of the proteome of thermophiles. We will use the information generated to 4) understand the in vivo stability of proteins, and 5) create genome-scale models of thermophiles to 6) optimize them as cell factories. Two main technological developments are thus expected:

- Instrumentation to determine the proteome of single prokaryotic cells and its response to temperature changes.
- Improved cell factory modelling by including proteome and thermal response information, so guiding metabolic engineering approaches to optimize environmentally sustainable industrial processes.

PROHITS represents a platform to train ten doctoral candidates (DCs) with translational and interdisciplinary, as well as transversal and entrepreneurial skills , resulting in:

- Excellent understanding of both experiment and computation, and of the challenges and opportunities at the interface between these.
- The ability to solve challenges in the determination, (biological) interpretation, and (industrial) application of thermophilic proteomes, along with broader method applicability to all prokaryotes.
- Engagement with the societal impact of their research, and awareness of innovation opportunities.

The Network is organised into six Work Packages as follows:



For more information, visit https://prohits.eu.

Consortium

PROHITS brings together 7+1 beneficiary institutions from six different European countries. Four academic organisations (VUB, VIB, UiB, UniVie, CNRS and UD) and two companies (NovoArc GmbH and Cellenion SASU) join forces with the support of two partner universities (Universiteit Gent and Université de Strasbourg) and a partner company (Bruker Daltonics GmbH& Co. KG) to build up a highly interdisciplinary network to tackle the ambitious goals of the project.

Partner institutions

Vrije Universiteit Brussel (VUB)
Prof. Dr. Wim Vranken
Prof. Dr. Eveline Peeters

Vlaams Instituut voor Biotechnologie (VIB) and Universiteit Gent (UGent)
Prof. Dr. Lennart Martens

Universitetet i Bergen (UiB)
Prof. Dr. Harald Barsnes

Universität Wien(UniVie)
Prof. Dr. Jürgen Zanghellini

Centre National de la Recherche Scientifique (CNRS) and Université de Strasbourg (UNISTRA)
Dr. Christine Carapito

NovoArc GmbH
Dr. Julian Quehenberger

Cellenion SASU
Dr. Anjali Seth

Debreceni Egyetem
Prof. Dr. Éva Csősz

Bruker Daltonics GmbH& Co. KG
Mr. Pierre-Olivier Schmit

Meet the DCs

Alexandre Bouillon

VUB VRIJE UNIVERSITEIT BRUSSEL

Alexandre has nurtured a lifelong passion for studying life in all its forms since his childhood in France. This curiosity, coupled with an early discovery of programming in high school, led him to pursue an engineering degree at EPFL in Switzerland. He earned both his Bachelor's and Master's in Life Sciences Engineering, where he honed a diverse skill set in molecular and cellular biology, mathematics, statistics, and artificial intelligence. During his time at EPFL, Alexandre engaged in numerous projects, including linking retinal features to the development of specific diseases, modeling the impact of cognition on fruit fly evolution, and validating lipidomics models for cell state prediction using computer simulations.

His work reflects a deep commitment to advancing our understanding of biological systems through computational and mathematical approaches. Alexandre's research interests encompass modeling, machine learning, computational biology, and the study of non-human organisms. Driven by a desire to push the boundaries of knowledge, he is dedicated to exploring the intricate mechanisms of life and contributing to the scientific community.

Josephine Boel Andresen

VUB VRIJE UNIVERSITEIT BRUSSEL

Josephine is of Danish-Norwegian descent and received her B.Sc. in Biology at the University of Copenhagen (UCPH) (DK), which included a study abroad to Imperial College London (UK) and a deep-sea expedition to the Sognefjord (NO) to study life in extreme environments. Developing an interest in the application of biotechnology for climate change mitigation, she completed her M.Sc. in Molecular Biology & Genetics at UCPH with a thesis focusing on photosynthesis-directed optimization of microalgae as biofactory units by investigating carotenoid-synthesising genes.

Afterwards, she was employed as a Research Assistant at the Technical University of Denmark to develop a biohydrogen production pipeline from strictly anaerobic and thermophilic bacteria isolated from the subsurface, in which she established laboratory procedures for a cultivation strategy with demanding circumstances

Caroline Jachmann

VIB

Caroline Jachmann completed her B.Sc. in Computer Science at the Heinrich Heine Universität Düsseldorf, diving deeply into data analysis and programming. Her fascination with the potential of combining biological problems with computational power led her to pursue an M.Sc. in Bioinformatics at the University of Tübingen. During her master's studies, Caro developed a significant interest in proteins, particularly through working with them in 3D visualisers for drug design applications during her master thesis.

To complement her computational skills, Caro undertook an internship at a proteomics facility, gaining hands-on experience in the wet lab environment. She is responsible for adapting the state-of-the-art tools from the CompOmics lab to the challenges of prokaryotic single cell proteomics. Outside the lab and office, Caro enjoys karaoke, running, volleyball, taking care of her thunder lizards, and propagating plants.

Kenneth Valerio Aguilar

UNIVERSITAS BERGENSIS

Kenneth, originally from Costa Rica, obtained his bachelor's degree in biology with an emphasis in biotechnology from the National University of Costa Rica in 2020. During his undergraduate years, he developed a passion for data analysis and bioinformatics. He worked as a student assistant in the Laboratory of Applied Bioinformatics (LABAP), where he gained extensive bioinformatics knowledge by actively engaging in various projects.

Immediately after graduating, Kenneth secured a position as a data analyst at a cloud computing consulting company. Here, he expanded his expertise in the latest cloud computing technologies and data management methods. In 2022, he moved to Hungary to pursue a master's degree in molecular biology at the University of Debrecen (UNIDEB). During his time at UNIDEB, he focused on the genotyping and analysis of genomic data from celiac disease patients, aiming to elucidate the main genetic factors and patterns associated with the disease. Currently, he aims to develop innovative methods for processing bioinformatics data and applying it to cutting-edge research.

María José Rimón Martínez

universität wien

María José Rimón Martínez studied Biology at the University of Murcia, Spain, where she gained extensive lab experience through internships focused on genomics and molecular biology. Her interest in these fields led her to conduct her Final Degree Project on using zebrafish as an animal model for psoriasis drug discovery, which sparked her interest in bioinformatics.

To deepen her expertise, María José pursued a Master's degree in Computational Biology at the Universidad Politecnica of Madrid. There, she honed her skills in several programming languages, applying them to modeling, lead discovery, and genomic analysis. For her master's thesis, she spent six months at the CNB (National Biotechnology Centre) collaborating with the Scientific Computing Service to design an R package called MADFBA (Multi-model Adaptive Dynamic Flux Balance Analysis) aimed at improving metabolic network simulations. María José has experience in both experimental and computational fields and is eager to continue learning and expanding her expertise.

Dinu Ziovie Ciobanu

cnrs

Dinu received the Master's degree in Chemical Sciences at University of Genoa. He completed the thesis in collaboration with University of Basque Country working on organic synthesis of bio-active compound via multi-component organocatalyzed reactions. After a short experience as Field Service Engineer, he joined IIT (Italian Institute of Technology) as Research Fellow working on two different projects: Graphene Flagship and FFC1.2023. In both projects he was involved in investigating the omics profiles, mainly proteomics and lipidomics, via mass spectrometry.

Dinu joins the PROHITS project aiming at conducting collaborative and interconnected research on proteomics studies on thermophiles. TPP mass spec based proteomics experiments will be conducted on bacteria, gathering information about protein stability both in bulk and at single cell level. Dinu joins as DC6 in a highly collaborative and interdisciplinary role with the final aim to generate, collect and output high quality proteomics data. This contribution is designed for the implementation of efficient and green industrial processes bacteria as biomass converters.

Meet the DCs

Sunu Lama

Sunu has been an Erasmus Mundus scholar of the joint master's degree program "EACH: Excellence in Analytical Chemistry" at the University of Tartu, Estonia (Master 1) and the University Claude Bernard Lyon 1, France (Master 2). She holds a bachelor's degree in pharmacy from Purbanchal University, Nepal and has gained professional experience as a pharmacist at Ohm Pharmaceutical Laboratories in Nepal. During her master's studies, she completed a six-month internship at LABERCA, ONIRIS, France in the "Alimentation" department under the Mixed Research Unit. Her research focused on analysing oligoesters and monomers in food coating materials (FCMs) and their gastrointestinal (GI) fates to ensure food's chemical safety, utilising LC/GC coupled with an Orbitrap high-resolution mass analyser. This experience has been a significant highlight in her academic journey where she garnered a wide range of research and analytical skills. Additionally, an opportunity to collaborate with the pharmacy researchers team of the University Santiago de Compostela, Spain, during the same project deepened her understanding of in vitro GI digestion and the protocols employed to investigate the same. Furthermore, her master's first-year internship at the Estonian Environmental Research Centre (EERC), Estonia specialising in "environmental chemistry", was another invaluable experience. With these professional experiences, spanning various laboratories encompassing both "analytical chemistry" and "pharmacy", amalgamated with her academic foundations, now her goal is to apply these analytical experimental and computational approaches to study molecular biology and leverage this knowledge to promote and enhance efficient pharmaceutical drug delivery systems. By participating in this promising scholarly research project "PROHITS", she aspires to delve deeper into single-cell proteomics, uncovering its hidden dimensions and making a meaningful contribution to this scientific community.

Ana Carvalho

Ana began her studies with her bachelor's in biotechnology at the University of Aveiro, in Portugal, in 2018. During these years, she developed an interest in microbial biotechnology and the bioprocessing field. Later in 2021, she enrolled in the master's in biotechnology at the same university, where she opted to do her master's thesis in the industry field, where she focused on the replacement of the current carbon source with a cheaper and greener alternative for a specific bioprocess, aiming to reduce production costs.

After finishing her studies, she enrolled in a 9-month Erasmus + internship at IPLA-CSIC, in Spain, studying the physiology, genetics, and technological properties of lactic acid bacteria that produce neuroactive compounds. There, she developed her hands-on skills in molecular biology techniques, bioinformatics, and microbial cultivation. In addition to science, she enjoys reading, sports, learning new languages, and playing trumpet.

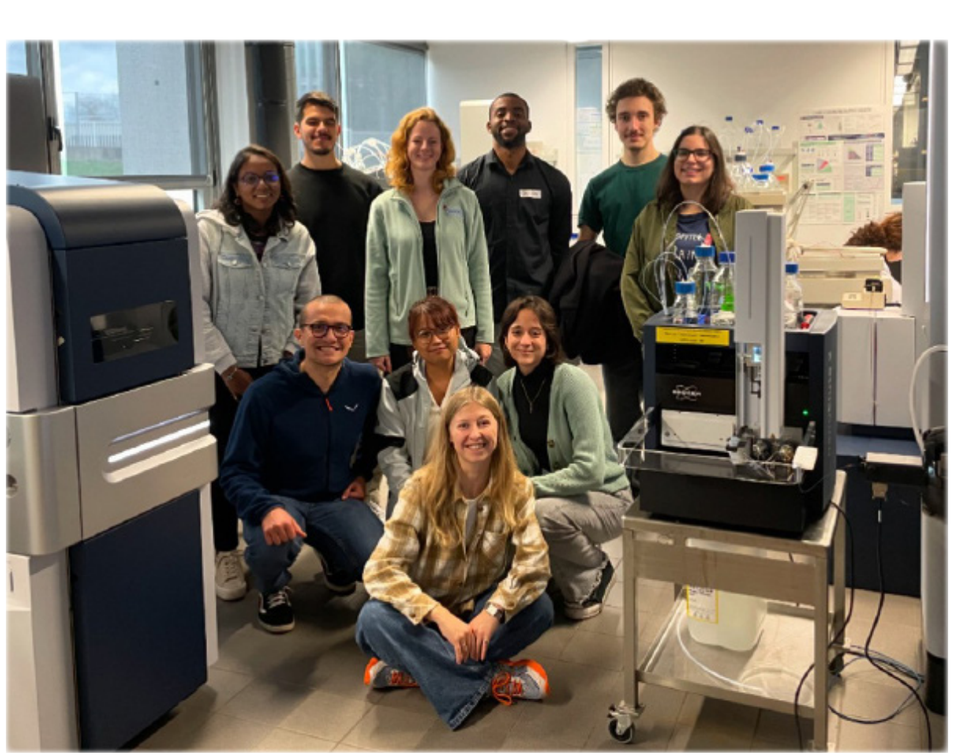
Onyeka Francis Offor

Onyeka Francis Offor was born in Nigeria where he obtained his bachelor's degree in pure chemistry at University of Port Harcourt (Uniport) Nigeria. After his graduation, he worked as a graduate assistant in the department of chemistry, Akwa State University, Nigeria. Thereafter he was awarded a fully funded scholarship for a master's degree in organic chemistry by the petroleum development technology fund (PTDF) at Uniport. His master's thesis at Uniport was on synthesis and modification of alkyd resin from velvet tamarind which also involved IR, GC-MS, and NMR characterization of the synthesized product. In 2022, following his completion of the master's program at Uniport, he was awarded a fully funded Erasmus Mundus Scholarship and proceeded to study in Europe for a dual master's degree in Analytical Chemistry at University of Tartu, Estonia and Uppsala University, Sweden.
During his second master's degree in Europe, he worked as a summer intern with Recipharm, Sweden where he developed LC-MS method for analysis of lipid (lipidomic). In June 2024, he completed his dual master's degrees in Analytical chemistry with a master's thesis focus on MALDI mass spectrometry analysis of acetyl-CoA and other CoA's relevant to humans (metabolomics). Francis is passionate about the omics research which has led to his involvement in the PhD research on proteomics.

Mansi Jain

Mansi was born in Delhi, the capital of India. She obtained a B.Sc. degree in Biochemistry from the University of Delhi. She was awarded the prestigious INSA-IASc -NASI Summer Research Fellowship during her undergraduate studies and further completed her master's at the University of Delhi, South Campus. During her master's thesis, she worked on protein purification techniques, which strengthened her research interest in the field of proteomics.

She then joined a mass spectrometry-based company in Delhi, where she was primarily involved in sample preparation and gained hands-on experience with MS instrumentation. Apart from her strong interest in the field of proteomics, she enjoys traveling, trying different cuisines and socializing with people.



Meet the supervisors

Prof. Dr. Ir. Eveline Peeters

VRIJE UNIVERSITEIT BRUSSEL

Eveline Peeters holds a PhD in Bioengineering Sciences and currently works as an Associate Professor in the Department of Bioengineering Sciences at Vrije Universiteit Brussel. Since 2014, she leads the Research Group of Microbiology, which is a dynamic research team specializing in molecular and synthetic microbiology. Eveline has always been affiliated with VUB, from her bachelor/master degree as a bioengineer until today. Her early academic journey at VUB was driven by a passion for extremophilic archaea, which are prokaryotes that thrive under extreme conditions. Under the mentorship of Daniël Charlier, she focused on studying into protein–DNA interactions for selected transcription factors and mechanisms of transcription regulation. After completing her PhD, Eveline enriched her expertise as a postdoctoral researcher through research stays at institutions like Rijksuniversiteit Groningen, Université Paris-Sud and Institut Pasteur. Research today spans from fundamental insights into transcription mechanisms to application-focused synthetic biology and biomaterial research. Her team investigates microbes across all domains of life, archaea, bacteria and fungi, with a final aim to engineer and optimize microbial cell factories for the sustainable production of materials and chemicals. Inter- and transdisciplinary research approaches are used. In addition to scientific leadership, Eveline is committed to high-quality education and cultivating an inclusive environment where research and training are closely integrated.

1. Describe your background, your research, your goals and contributions to the field
I am a bioengineer by training and have always been affiliated with VUB in Brussels, Belgium (as a student, PhD researcher, postdoc researcher and now as professor). My goal is to contribute to advancing our understanding of molecular processes in microorganisms and to developing new biotechnological applications with microorganisms. My past contributions were mainly focused on the study of gene regulatory processes in archaea, with as a model organism Sulfolobus.

2. Present your research group
The VUB research group of Microbiology is a research group with a broad mission statement: we aim to better understand molecular mechanisms of stress response in microorganisms and to implement synthetic biology approaches for the engineering of microbes for bioprocesses for production of chemicals and materials. The research activities within the group are organized to create synergies between fundamental and application-driven research lines, as well as to enable the use of inter- and transdisciplinary approaches. Equally important in the research group's mission statement is that we strive to create an inclusive research environment, in which all researchers are empowered to further develop their talents and pursue their interests.

3. What inspired you to pursue a career based on research?
Not "what" by rather "who": my previous mentor and PhD promoter Daniel Charlier. He taught me that research is a bottom-up process, driven by curiosity and creativity, but also guided by a humanistic perspective. This resonated so much with me that it inspired me to further pursue a research career.

4. What has been a significant moment in your research path?
The start of my trajectory as an assistant professor (back in 2014): at that time, I realized how my fundamental research interests - studying extremophiles and archaea - can be linked to application-oriented developments that assist in the transition to a more sustainable and environmentally friendly industry and society and it marked the foundation of the research group as it is today.

5. What outlines a good supervising and/or management strategy in your eyes and how do you implement it in your research group?
For the supervision of researchers, I use a coaching approach, by being closely involved in the research projects but also stimulating each researcher to take charge of their own trajectories. On the level of the research group, we operate by creating an interplay between a sociocratic approach, in which all group members are involved in decision making processes and in which we build on our collective intelligence, and a top-down approach, where the management takes up responsibilities when needed by the team.

6. What does PROHITS mean to you?
The core concept of PROHITS embodies the close connection and collaboration between PROHITS coordinator Wim & myself, by being built on the interface and synergy by our research interests (thermophiles and proteomics), as well as our complementary research approaches (experimental and computational). As such, it is a project close to my heart. Now that the project is full swing, I thoroughly enjoy the scientific interaction with all co-PIs, as well as with all Doctoral Candidates, which all bring unique talents and perspectives.

7. What is your and your team's contribution to PROHITS?
We will contribute to the experimental and fundamentally oriented research goals in PROHITS. Building on our core expertise, we will develop cultivation procedures for different thermophilic strains, and aid with the biological interpretation of the proteomic data generated in the project.

8. Which milestones do you think are the most important?
An important milestone is when we will have established a complete pipeline from cultivation to the generation of high-quality proteome data for one of the thermophilic species.

9. What challenges might we expect in PROHITS?
For me, the biggest challenge will be to gradually make decisions on which biological questions to focus, given the broad initial scope and the big datasets that are expected to be generated.

10. What impact do you think PROHITS will have? Both within science but also societal?
Within science: unprecedented biological insights into biotechnologically promising prokaryotes, some of which have been understudied thus far. Within society: the interdisciplinary training of high-potential researchers within the PROHITS project will enable to generate impact in the field in microbial biotechnology in many ways: through further research trajectories or entrepreneurship, our PROHITS Doctoral

About - MICR at VUB

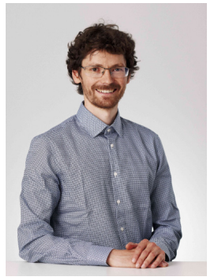
The Research Group of Microbiology (MICR) at Vrije Universiteit Brussel performs research in molecular microbiology and microbial biotechnology, aiming to deepen our understanding of the physiology and lifestyle of a broad spectrum of microorganisms, including archaea, bacteria and fungi, with particular expertise in extremophiles. Their investigations focus on unraveling molecular mechanisms underlying microorganisms' responses to environmental stresses and nutritional shifts, centering on gene regulation and protein–DNA interactions.

Utilizing interdisciplinary techniques such as genetics, genomics, and biochemical methods, the group bridges fundamental discovery with application:

- **Engineering microbial cell factories and synthetic-biology toolboxes to produce biobased chemicals**
- **Biomaterials like mycelium composites and bioplastics**
- **Contribute to a sustainable circular economy**

Based on VUB's Main Campus in Brussels, MICR engages in collaborative projects, also involving industrial partners, and plays an important educational role by training students in cutting-edge microbiology and synthetic biology.

Meet the supervisors



Dr. Julian Quehenberger

Dr. Julian Quehenberger began his academic training in chemistry at TU Graz, Austria, before shifting his focus to biotechnology at TU Wien, where he earned both his Master's and doctoral degrees in Technical Chemistry and Biochemical Engineering. During his Ph.D., he established Sulfolobus acidocaldarius as a novel host system for extremophile biotechnology. As a postdoctoral researcher, Julian focused on CO_2 fixation in Sulfolobales under extreme conditions. His scientific work spans cultivation, molecular biology, lipid mass spectrometry, and bioprocess development, with a strong emphasis on translating fundamental extremophile biology into industrial applications.
He is now the Co-Founder, Managing Director, and CTO of NovoArc GmbH, a TU Wien spin-off. At NovoArc, Julian leads the technological development of lipid-based excipients derived from archaeal sources, opening new frontiers in oral drug delivery and pharmaceutical formulation. His work has resulted in patents, peer-reviewed publications, and recognition through innovation awards across Austria and Europe, with a research focus on archaeal lipids in drug delivery and the physiology of S. acidocaldarius.
Julian also serves as a lecturer at IMC FH Krems, where he contributes to academic training in applied biosciences and biotechnology.

1. Describe your background, your company, your goals, and contributions to the field.
My name is Julian Quehenberger, and I am the co-founder and CTO of the biotech company NovoArc. I have an academic background in chemistry and biotechnology, with a focus on utilizing microorganisms for the production of value-added products. During my PhD research, I studied the extreme thermoacidophilic archaeon Sulfolobus acidocaldarius, aiming to harness and enhance its metabolic capacities to utilize carbon-rich, acidic waste streams from the pulp and paper industry for the production of the natural sweetener xylitol.
As my understanding of this unique organism deepened, my doctoral supervisor, a colleague, and I recognized the potential of its distinctive membrane lipids for drug delivery applications. Due to their unique stereochemistry and ether-based structure, evolutionary traits shaped by the archaea's ancient origins and extreme growth conditions, these lipids offer remarkable stability, protecting drugs during storage and administration while improving the bioavailability of active pharmaceutical ingredients (APIs).
Our most significant contribution, and the technological foundation of NovoArc, was the development of a scalable cultivation and purification process for these archaeal lipids. This breakthrough allows their production in the quality and quantities necessary for pharmaceutical applications. Our goal is to supply academia and industry with these specialized lipids, driving global research in this promising field while enhancing the effectiveness of existing and emerging drug formulations by combining high-end API development with an innovative approach to drug protection and delivery.

2. Present your company.
NovoArc was founded in 2021 as a spin-off from the Technical University of Vienna. Today, we are a team of 14 professionals, most with an academic background ranging from bachelor's degrees to postdoctoral levels. In addition to an investor, the university and public funding agencies played a crucial role in our foundation, providing support in business development, legal matters, and start-up financing through grants.
Our company is based in Vienna, where we operate our own office space and research facility. NovoArc specializes in cultivating extreme thermophilic archaea, lipid extraction and purification, and lipid chemistry. Additionally, our formulation department has extensive expertise in encapsulating APIs in liposomes and lipid nanoparticles. This department carries out in-house research as well as collaborative projects with pharmaceutical industry partners, aiming to improve both established and novel drug formulations.

3. What inspired you to pursue a career based on research?
I've always been driven by a deep curiosity about how biological systems function, particularly how organisms interact with and adapt to their environments. The ability to build upon existing knowledge, uncover new insights, and contribute, however small, to discoveries that future generations can expand upon is incredibly motivating. Research offers the unique opportunity to not only understand fundamental processes but also apply them in meaningful ways.

4. What has been a significant moment in your research path?
One of the most rewarding moments in my research journey was investing significant effort into a complex challenge, as a team formulating and developing multiple hypotheses along the way, and finally obtaining concrete evidence that validated one of them. A particularly memorable instance was when for the first time we held a vial containing several milligrams of an archaeal lipid of highest purity in our hands that we obtained from dried archaeal biomass. Seeing it in its isolated form and being able to observe and study it in all its beauty with the naked eye was a powerful and memorable moment, turning a very abstract concept into something tangible and ready for further exploration.

5. What outlines a good supervising and/or management strategy in your eyes, and how do you implement it in your company?
In research, I believe it's essential to allow room for exploration and unexpected discoveries by maintaining wide boundaries for creative freedom. At the same time, structured discussions within a team are invaluable, as they provide fresh perspectives and help refine ideas. At NovoArc, we encourage this balance, giving our team the flexibility to explore, while also fostering collaboration through regular discussions. As a company, we do face constraints such as resource availability and business priorities, but strategic public funding can help push those boundaries further and enable more fundamental research.

6. What does PROHITS mean to you?
PROHITS represents an incredible opportunity to connect with highly motivated students and principal investigators from diverse fields from all over the world. It enables interdisciplinary exchange, provides fresh insights, and supports the next generation of scientists. More broadly, it strengthens research collaboration within the EU, ensuring that talent is nurtured and innovation continues to thrive.

About - NovoArc Gmbh

NovoArc GmbH is an Austrian biotech SME founded in 2021 by David Wurm, Julian Quehenberger, and Oliver Spadiut. The company develops and produces unique tetraether lipids (TELs) that serve as novel, biologically derived drug delivery systems. Using a patented, continuous bioprocess and extremophilic microorganisms, NovoArc manufactures these lipids in a scalable, sustainable, and highly reproducible manner at its dedicated research and production facility in Vienna.

TELs offer exceptional acid stability, heat resistance, and oxidation protection, allowing for oral delivery of sensitive drugs without chemical stabilizers and improving bioavailability. In vitro and in vivo studies confirm their non-toxicity and performance advantages.
NovoArc operates as a B2B R&D-driven company, with a business model based on:

- **Lipid sales in mg to g scale to industry and academia**
- **Feasibility studies for drug formulation**
- **Joint development projects with industrial partners**

NovoArc currently employs 13 staff members and targets the novel drug delivery market, particularly oral therapeutics, combining commercial traction with deep scientific innovation.

Meet the supervisors

Continued from the previous page

7. What is your and your team's contribution to PROHITS?
Our contribution to PROHITS is bringing an industrial and applied perspective to the research. While fundamental science is essential, translating discoveries into real-world applications comes with its own set of challenges. We provide insights from a production and scalability standpoint, helping bridge the gap between academic research and industrial implementation.

8. Which milestones do you think are the most important?
Developing predictive models that enhance mechanistic understanding is a crucial milestone. If we can accurately model and predict biological behaviors, it will enable targeted experimental adjustments, making research and application development much more efficient and precise.

9. What challenges might we expect in PROHITS?
As with any large collaborative research initiative, coordination and interaction across multiple disciplines and institutions can be challenging. However, PROHITS seems to have an excellent structure and administrational team to manage this effectively. On the technical side, single-cell analytics presents significant challenges, particularly in terms of sensitivity, resolution, and data interpretation. Overcoming these hurdles will be key to the project's success.

10. What impact do you think PROHITS will have? Both within science and society?
From a scientific perspective, PROHITS will advance our understanding of extremophiles and their biotechnological potential. It will provide valuable mechanistic insights into model organisms and help develop novel applications. On a societal level, the knowledge gained could lead to innovations in biotechnology that address real-world challenges, from pharmaceuticals to sustainable processes. Additionally, by nurturing new talent, PROHITS will contribute to both academia and industry by shaping the next generation of scientists and innovators.

DCs in Brussels, April 2025

Project progress

Meetings

Kick-off meeting
The kick-off meeting took place on 30th January 2024, counting with the participation of all beneficiaries and representatives of partner universities. During the meeting, the members of the supervisory board and the subcommittees were appointed, and the main practical organisation and management topics were discussed and agreed on.

Recruitment meeting
The recruitment meeting took place in Brussels on the 15th of May 2024 and was preceded by an 'ice-breaker' socializing event the evening of the 14th of May. Thirty-two candidates were invited to the event; 19 of them attended the meeting in person and the other 13 were present virtually.

Mid-term check
The PROHITS mid-term check meeting took place in Brussels on 8th April 2025. The Project Officer joined the Consortium to discuss the 1st year of project implementation and meet the 10 DCs.

Supervisory Board and subcommittee meetings
The Supervisory Board meets twice per year to discuss all high-level project matters, and has Caroline Jachmann (DC3) as DC representative.
The research subcommittee meets online once per month to regularly discuss key scientific topics. It involves all supervisors and DCs.
The data management, and dissemination and training subcommittees meet online, typically every 2-3 months. They count with the involvement of Kenneth Valerio Aguilar (CD4) and Josephine Boel Andresen (DC2) as DC representatives, respectively.

Deliverables and milestones completed so far

Deliverables

D5.1 Advertising of all DC positions
D2.1 Supervisory Board of the Network
D6.1 Project website and social media networks online
D5.3 Signed document with common IP strategy
D4.1 Career Development plans
D5.4 Progress report
D5.5 Data management plan
D6.2 Plan for dissemination and exploitation of results, including communication activities
DX.Y Lorem ipsum

Milestones

M18 Consortium Agreement
M19 Planned recruitments completed
M20 All recruited fellows enrolled in PhD programme
M21 Project mid-term check

Project progress

Workshop 1 - Project kick-off and DC initiation

The very first PROHITS workshop took place in Strasbourg on 7th-11th October 2024 in Strasbourg (France), with CNRS acting as host. The Consortium kickstarted the meeting with a detailed project overview presented by Prof. Wim Vranken that was followed by the DCs' introductory presentations. Such presentations helped DCs already identify connections with peers throughout the project to jointly answer key scientific questions. The whole consortium gathered in a social evening before the DCs embarked in their first trainings, with included:

- **Introduction to Mass Spectrometry and Proteomics,** led by Prof. Laurence Sabatier
- **Python/Jupyter Programming for Bioinformatics,** led by Caroline Jachmann (DC3)
- **Presenting with impact,** led by Hans Van de Water (The Floor is Yours)

DCs getting a tour of the Mass Specrometry laboratory by Dr. Christine Carapito at CNRS

A non-scientific team building activity was also completed to strengthen the bonds of the newly starting DCs. The DCs split in two groups to solve virtual challenges.

Group picture of DCs awaiting their virtual reality game assignments

Mid-term check + Workshop 1.5

The PROHITS consortium met again in early April 2025 in Brussels. The mid-term check meeting with the Project Officer from REA was held on 8th April, and was followed by a newly planned Workshop 1.5 (9th-11th April).

The PROHITS Consortium during the mid-term check with the Project Officer from REA

The event included a meeting of the Supervisory Board, DCs' progress presentations, a team building activity for DCs and the following training sessions:

- **Thermal Proteome Profiling,** led by Dr. Christine Carapito
- **Principles of Open Science and FAIR data,** led by Prof. Lennart Martens
- **Laboratory practical on thermophile biology,** led by Josephine Boel Andresen (DC2)
- **Science Outreach,** led by Sven De Boeck (VUB's Science Outreach office)

DCs in the laboratory of MICR at VUB

DCs enjoying the nice weather between meetings

Project progress

News

- Caroline Jachmann (DC3) gave a workshop entitled “Fanatastic PTMs and how (not?) to find them using msqrob2PTM - a real-life journey” at the EuPA-FPS 2025 conference (16-20th June 2025 in Saint-Malo, France).
- Josephine Boel Andresen (DC2) volunteered at the Soapbox Science festival (28th June in Brussels, Belgium)
- Alexandre Bouillon (DC1) and Jospehine Boel Andresen (DC2) will participate in the I Love Science Festival coming to Brussels (12-13th October 2025) with a wet lab protein booth called “Protein to the extreme!”. The festival is schools-only Friday and open to all during the weekend.
- The next PROHITS workshop will this time take place at the University of Debrecen in Hungary, 24-28th November 2025.

Caroline (DC3) at EuPA.

Contributions at scientific meetings

Andresen, J. B., Peeters., E. Thermophilic proteomics - Turning on the heat for a new generation of microbial cell factories. Poster presented at the Belgian Society for Microbiology’s Annual Symposium 2025, 21st February 2025, Brussels (Belgium).

Jain, M., Carvalho, A., Quehenberger, J., Andresen, J. B., Peeters, E., Csosz, E. Assessment and Comparison of Sample Preparation Strategies for High Yield Proteomics Analysis of Thermophilic Prokaryotes. Poster presented at the Hungarian Molecular Life Sciences 2025 Conference, 28-30th March 2025 (Hungary).

Carvalho, A. C., Peeters, E., Zanghellini, J., Quehenberger, J. Towards a novel tetraether lipid producer Haloferax volcanii. Poster presented during the Panel B PhD retreat of the Vienna Doctoral School in Chemistry (DoSChem) of University of Vienna, 2nd April 2025 (Austria).

Rimón, M. J., Zehetner, L., **Zanghellini, J.** From genome to metabolism: Pan genome-scale metabolic model reconstruction of the extremophile genus Sulfolobus. Poster presented during the Panel B PhD retreat of the Vienna Doctoral School in Chemistry (DoSChem) of University of Vienna, 2nd April 2025 (Austria).

Jain, M., Carvalho, A., Quehenberger, J., Andresen, J. B., Peeters, E., Csosz, E. Evaluation of Different Sample Preparation Strategies to Identify the Best Approach for High-Yield Bottom-Up Proteomics Analysis of Thermophilic Prokaryotes. Poster presented at the European Proteomics Association 2025 Conference, 16-20th June 2025 (France).

Valerio-Aguilar, K., Barnses, H. Cracking the Heat Code: Proteomic Adaptation in Thermophilic Microbes. Poster presented at the Bioinformatics in Bergen 2025 Conference, 2-3th June 2025 (Norway).

Bouillon, A., Vranken, W. Do you speak protein? Understanding and Predicting Protein Thermal Stability. Poster presented at the Bioinformatics in Bergen 2025 Conference, 2-3th June 2025 (Norway).

Andresen, J. B., Jain, M., Peeters, E. Turning up the heat in temperature-responsive gene regulation of a thermophilic cyanobacterium. Poster presented at the 15th Workshop on Cyanobacteria, 4-7th June 2025 (USA),

Journal club

The fellows meet online every 2-3 months to discuss about articles relevant to the project.

1st Journal club session:

Date: 17 December 2024
Article: Transcriptional and translational dynamics underlying heat shock response in the thermophilic crenarchaeon Sulfolobus acidocaldarius
DOI: https://doi.org/10.1128/mbio.03593-22
Chair: Josephine Boel Andresen (DC2)

2nd Journal club session:

Date: 18 February 2025
Article: Toward Single Bacterium Proteomics
DOI: https://doi.org/10.1021/jasms.3c00242
Chair: Onyeka Francis Offor (DC9)

3rd Journal club session:

Date: 15 April 2025
Article: Meltome atlas—thermal proteome stability across the tree of life
DOI: https://doi.org/10.1038/s41592-020-0801-4
Chair: Alexandre Bouillon (DC1)

4th Journal club session:

Date: 1 July 2025
Article: Oxidative Stickland reactions in an obligate aerobic organism – amino acid catabolism in the Crenarchaeon Sulfolobus solfataricus
DOI: https://doi.org/10.1111/febs.14105
Chair: Ana Carvalho (DC8)

Secondments

Completed:

DC8 Ana Carvalho at **VUB, Belgium**.	Nov 2024 - Feb 2025	Duration: 4 months
DC6 Dinu Ciobanu at **Bruker, Germany**	March 2025	Duration: 1 month
DC7 Sunu Lama at **Bruker, Germany**	March 2025	Duration: 1 month
DC9 Onyeka Francis Offor at **Bruker, Germany**	March 2025	Duration: 1 month
DC2 Josephine Boel Andresen at **CRNS, France**	May 2025	Duration: 1 month
C3 Caroline Jachmann at **CNRS, France**	May - June 2025	Duration : 1.5 months

Ongoing:

DC4 Kenneth Valerio at **VIB/UGent, Belgium**	July 2025	Duration: 1 month

Stay tuned!

Linkedin
https://tinyurl.com/4cayu96f

PROHITS newsletter
https://tinyurl.com/3wesyhv3

Bluesky
https://tinyurl.com/535s4s9f

Instagram
https://tinyurl.com/2a6vssjc

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