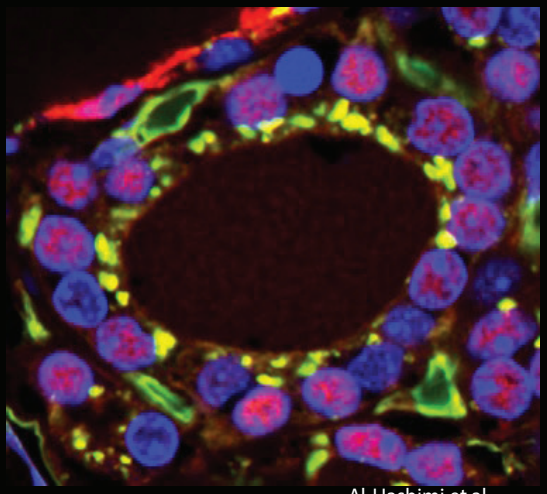
July 2024

IN THIS ISSUE:

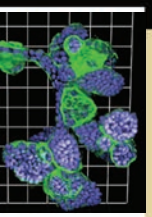


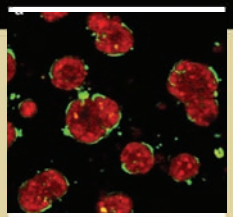
Al-Hashimi et al.
Biochim Biophys Acta
Mol Cell Res. 2020

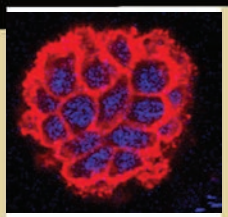
INTERNATIONAL PROTEOLYSIS SOCIETY

QUICKCUTS

Edited by:
Laura Edgington-Mitchell







THE PREMIER RESOURCE
FOR ALL YOUR IMPORTANT PROTEASE NEWS

IPS COUNCIL

A Message From the President:

Dear IPS community, dear friends

It's been seven months since our last QuickCuts edition, and we have several updates to share.

As announced in the previous QuickCuts, we mourn the loss of two cherished IPS members: Bonnie Sloane, co-founder and first president of the IPS, and Margarete Heck, a very active and supportive IPS member. This edition includes an obituary honoring their contributions. Thank you to everyone who shared their memories and tributes. They are deeply missed.

I wish to express my gratitude to the anonymous IPS members who made private donations to establish the Heck-Sloane Fellowship Award 2024, in memory of Bonnie and Margarete. Your generosity funded 10 fellowships for trainees to attend the recent GRC on Proteolytic Enzymes & their Inhibitors, embodying their spirit of mentorship. Thank you for your support.

In the spirit of promoting trainees, the IPS also awarded Nicholas Young (UCSF) with the Best Presentation Prize at the GRS. Congratulations, Nicholas!

Meanwhile our webinar series continues to thrive. Recently, we enjoyed an insightful talk by Irit Sagi on 'Exploring "ECM Remodeling Memory" as a Mechanism for Damage Accumulation in Acute and Chronic Inflammatory Diseases.' Thank you, Irit! The sixth webinar is scheduled for September, and will be delivered by Guy Salvesen, entitled 'Being a Scientific Mentor - Lessons I Learned from My Mentees.'

This QuickCuts edition shows our society is flourishing, thanks to the strength and dedication of its members. I encourage you to renew your membership and stay in touch with us, the IPS council, to provide your feedback and suggestions. Importantly, preparations for the 13th IPS General Meeting are ongoing. Ana Paula Lima and Maria Luiza Oliva are working on a fantastic program that you do not want to miss. Mark the date in your calendars: October 26-30 in Búzios, Brazil.

Finally, I extend my heartfelt thanks to thank Laura Edgington-Mitchell for her excellent work in editing this issue of QuickCuts.

Best wishes

Ruth Geiss-Friedlander
Email: ruth.geiss-friedlander@mol-med.uni-freiburg.de

Announcing IPS 2025

Chaired by Dr Ana Paula C. A. Lima

Early Career Workshops will be held ahead of the meeting at Federal University of Rio de Janeiro

Save the date for our 6th IPS Webinar

Prof Guy Salvesen
Sanford Burnham Prebys

Prof. Guy Salvesen is a South African-born biochemist, best known for his work in the field of apoptosis. His research focuses on proteases and their inhibitors in humans, with particular emphasis on the caspases of the apoptotic cell death pathway. His PhD in biochemistry is from the University of Cambridge, studying under Alan Barrett (1981). His first posts were at the Strangeways Research Laboratory and MRC Laboratory of Molecular Biology in Cambridge. In 1985, Salvesen moved to the USA, taking up a position at the University of Georgia. He joined the faculty of Duke University in 1987, and moved his laboratory to the Sanford-Burnham Institute for Medical Research, La Jolla, California in 1996. For his work, he received several academic prizes. Guy Salvesen was a mentor to many Ph.D. students and Post-docs. He is also a co-founder of International Proteolysis Society.

Please save the date for Guys webinar:

Wednesday 25th September 2024
8:00 am PDT
5:00 pm CEST

Register for this virtual event at
https://www.protease.org/ips-webinars

In Memoriam: *Dr Bonnie Sloane*

Professor Bonnie Sloane was born in Pittsburgh, Pennsylvania on August 12, 1944. She received her bachelor (1966) and master (1968) degrees from Duke University and a PhD from Rutgers University in 1976. Her first academic post was at the University of Pennsylvania as a postdoctoral fellow. She then moved to Michigan State University as an Assistant Professor in 1979 and ultimately to Wayne State University in the Department of Pharmacology in 1980. She was promoted to associate professor in 1984 and to professor in 1989. In 1996, she was elected to the Wayne State University Academy of Scholars and in 2005 she was named Distinguished Professor. She was the Pharmacology Chair from 1995 to 2015, the first woman to serve as chair of a department in the Wayne State University School of Medicine. In 1999 she co-founded the International Proteolysis Society and was elected as its first president. From 2009 until 2011, Professor Sloane was the president of the Association of Medical School Pharmacology Chairs, again the first woman to serve in this role, and in 2021 was named a fellow of the American Association for the Advancement of Science.

Professor Sloane was recognized by her peers as a visionary, a pioneer and an extraordinarily dedicated scientist and educator in the field of proteolysis in cancer. She was the first to propose that the lysosomal proteases are secreted in cancer and play a pivotal role in tumor progression and metastasis. She was internationally known for her innovative imaging-based approach for a model system to study functional proteolysis in tumor microenvironment interactions in 4D (3D+time). Based on this work, in 2002 she established the Breast Cancer Center of Excellence (BCCOE) at WSU with funding from the Department of Defense. The BCCOE was a multi-institutional project which quickly became a magnet for attracting international scholars. As such, her laboratory and WSU became the go-to-center for training. Numerous students, fellows as well as established scientists from every corner of the world came to her laboratory to receive training. She leaves behind an outstanding scientific track record and an enviable legacy that is recognized and respected internationally.

Kamiar Moin, Wayne State University

For those who don't know me, I was a member of Bonnie's original research group and was with her longer than anyone else, for almost my entire professional career. In this regard you might say that I knew her better than anyone outside her immediate family.

I first met this remarkable woman in spring of 1986 when she interviewed me for a postdoctoral position in her laboratory. I started working with her the following October and remained at her side for the next 37 years. She was my mentor, my professional partner, and above all my best friend and confidant. Here, however, I don't want to write about her outstanding professional career and achievements which most of you in this forum are already aware of. Instead, I want to write about her wonderful character and what made her so special. One of Bonnie's most outstanding and important qualities was her ability to recognize one's strength and potential in short order. She would then provide guidance and a gentle push so that one could focus on that strength and achieve one's full potential. She was a role model for so many young scientists, particularly women including my own two daughters.

Bonnie's core belief was that students, postdoctoral fellows and young faculty who are starting their careers need recognition and the opportunity to show their work. To that end, she never spoke in the sessions that she chaired. Instead, she always had students and trainees, whether from her group or others, present their work. She had her students and postdocs accompany her to most conferences that she attended, and not just one or two but, often 3-5 of them, sometimes the entire laboratory. On one such occasion, on a lighter side, her group had such prominent presence by sheer numbers present that our colleague Jim McKerrow referred to Bonnie and her group in his presentation as the Detroit mafia! She introduced her students to prominent scientists and made sure that they were noticed. Her relentless conviction to the welfare of her students and trainees was the reason she was so revered by them. And this did not go unnoticed. Everyone wanted to know the nature of this special relationship. On one occasion, at the IPS General Meeting in Portoroz, I was approached by a student from another group who wanted to know the reason why Bonnie's students seemed so happy, to which I pointed at Bonnie and said she was the reason. Because of this special relationship with her students as well as the pioneering work that was being done by her group, her laboratory became a magnet to attract students and scholars from all around the world. Every single continent at some point or another was represented in her group. Students, postdocs and even young faculty from over 21 countries received training in Bonnie's laboratory through the years. They came not just to receive training in groundbreaking work, but also to experience this special relationship.

In closing, I always considered myself as a pragmatic and a resilient man. However, no degree of pragmatism or resiliency can heal the scars that Bonnie's passing has left on my heart. I miss her so!!

Klaudia Brix, Bremen

Distinguished Professor Dr. Bonnie F. Sloane was an exceptional scientist with the most generous and supportive personality that I could imagine. It is an honor to call her my role model and my dear and close friend!

The first time I met Bonnie was at a meeting in Panama City Beach. I was new to the field and had just published my first paper on cysteine cathepsins in the thyroid gland. It was important to be at the meeting to make contacts and get feedback. So, at one of the first evenings, I stepped out of the elevator as I was on my way to the room, while Bonnie as the co-chair of this conference stepped in, because she was on her way to the poster session. She looked at me, made sure not to be mistaken by reading my name tag, and then said "You did good work." This short Bonnie-style sentence was an encouraging invitation and decisive for me to continue in the field of proteases. Bonnie supported me in every step of my career as if I were one of her group members. She would always take the time to look at my posters or to attend my presentations, to comment and to give advice on how to proceed. Her comments were critical, to the point, and helpful at the same time. We met at many more conferences to discuss about cathepsin B et al.

Bonnie invited me over to Detroit in 1996, this was one of my first official Invited Speaker talks. I was so happy and already at this visit it became clear that Bonnie was perhaps also the best teacher to learn how to combine science and family. We went out for a fancy dinner, but of course, before going there, we needed to stop and see her husband and son, who was playing soccer. I was touched because it really meant she was serious about building a wonderful friendship. Bonnie was also the first who visited while I was still in Bonn, what an honor that she traveled with me, after a great but intense Winter School in Tiers, all the long way from Munich by train. She also came several times to Bremen, although this meant a lot of work in the university's advisory board and it interrupted her preparation for celebrations with her family. Then, I was perhaps not appreciating enough, how generous this was from Bonnie and Doug's perspective.

I will not forget when Bonnie showed up at a GRC in New England, the place with the frog pond. She could not wait to show me pictures of a lovely forest by the lake where she and her husband were planning to build a house. Bonnie would keep me updated throughout the building phase and it meant a lot to my architect husband Klaus and me to be invited to this spectacular and beautiful house. Then, we took our first selfie – no joking. It was another wonderful visit in Empire, in 2017, when Margarete and I were invited and shown around in the sleeping bear dunes by Bonnie, while Doug was preparing excellent dinners for us.

Bonnie, Margarete, Judith, Lakshmi and I had fantastic times before, at and after conferences. We traveled in South Africa, Slovenia and Italy. How enjoyable it is to think back to so many good memories! When the pandemic came along, we started a group chat that kept us all going as I wish to believe, it was and still is unique and very precious to me. We kept and keep going with our chats, remembering you, dear Margarete and dear Bonnie very fondly.

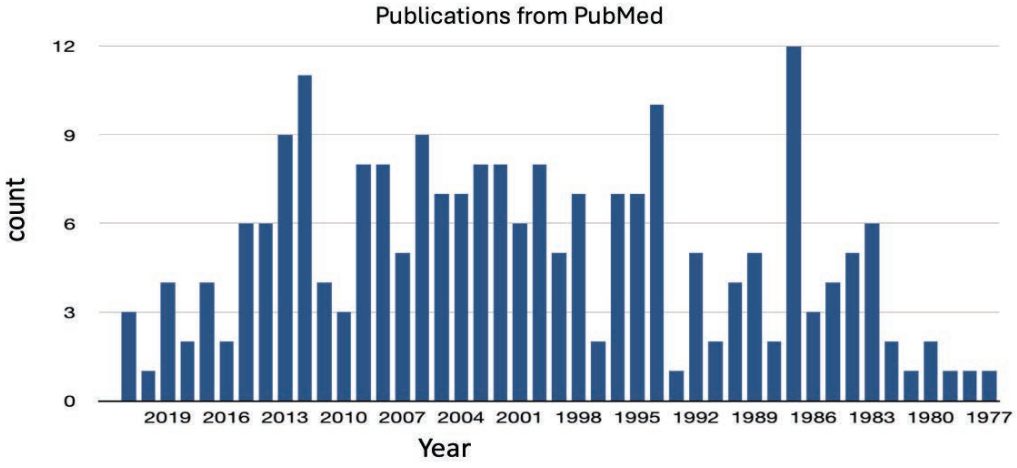
Bonnie, you are missed so much! Thank you for your friendship and love, for your science advice and endless support, for the fantastic times we could spend with you.

My deepest sympathies go to Doug, Becky and Stuart and your Grandchildren, and to your research family and friends in Detroit and all over the world.

Guy Salvesen, Sanford Burnham Prebys

I met Bonnie Sloane in a restaurant in New Orleans during a Federation of American Societies for Experimental Biology (FASEB) meeting. I think it was spring of 1981, but it could have been 1982, so don't quote me on this. As is sometimes the case, our lifelong friendship was inspired by a third party. Bill Schwartz, a postdoc in the lab where I was working on my doctorate, had been supervised by Bonnie's mentor John Bird at Rutgers and had told me that I should look up Bonnie if I ever had the opportunity. What better occasion than to meet up in one of the culinary hotspots of the country, especially given that Bonnie was somewhat of a gourmet. As many will remember, Bonnie was a quiet person with an engaging personality and I think this became key to our friendship, balancing my sometimes-boisterous view of life. Bonnie was a fantastic networker and organizer, working with Judy Bond to lead the 9th (and final) meeting of the International Committee on Proteolysis (ICOP) in Williamsburg, Virginia in October 1992.

Bonnie had an impressive publication rate, collaborating with many groups in the field. Her first paper was in 1977 and last one in 2021, and although we never published together, we kept in touch through our mutual interest in the universe's most intriguing enzymes (proteases) as we built our overlapping professional networks. With ICOP winding down, the field needed a different kind of organization to represent its interests, and it was with the urging of ICOP leaders that Bonnie and I joined together (again in a restaurant) in San Diego to plant the seeds of what became IPS. To Bonnie it was important that IPS provide a forum for junior investigators (postdocs and students) to hone their skills. I credit Bonnie with ensuring that IPS become an organization with a major focus on training the next generation of scientists, and I thank her for helping to instill this aspect into my own work habits. Bonnie's insistence on including junior investigators in oral sessions at various scientific meetings, including IPS, is something that we must all learn from to ensure that the next generation of scientists emerges with good communication and networking skills. It is this razor-sharp focus on training and mentoring plus her delight of all things culinary that rest peacefully in that part of my memory reserved for this beautiful friend and mentor.

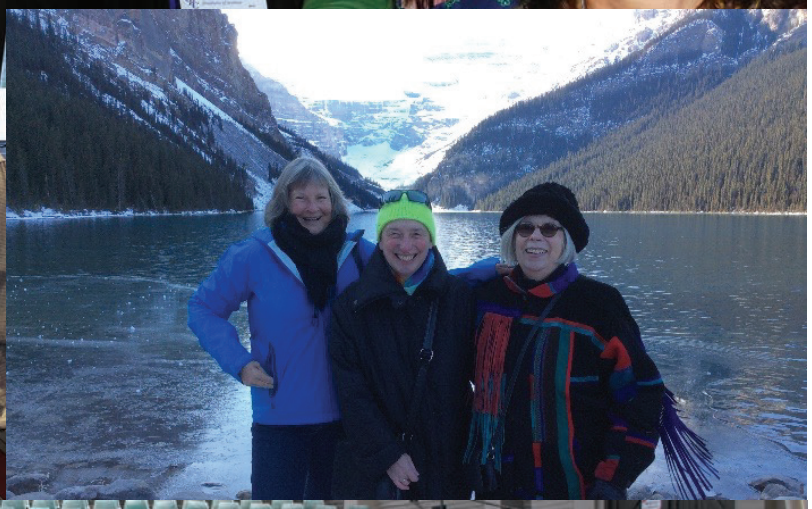


Judith Clements, Queensland University of Technology

I think that the first time that I met Bonnie was at an AACR meeting in San Francisco in 2002. It was a brief engagement, but I was really impressed with Bonnie's work and delighted to meet another scientist working on proteases in cancer. Over the years, attending Gordon Conferences, Winter Schools and IPS, we became good friends, and although not collaborators, given that we worked on different proteases and different cancers as well, I benefited from Bonnie's insightful comments on my work and the field in general. I particularly loved her work with Kamiar Moin on live cell imaging of cathepsin proteolysis in the 4D microenvironment of breast cancer as exemplified in their 2019 Cancer Metastasis Review. Such a terrific marriage of enzymic analyses in real time and what wonderful photo-microscopy of the breast cancer microenvironment. When I led a 'Proteases in the Tumour Microenvironment" mini-conference in Prato, Italy in 2017 and 2019, Bonnie was a key participant – and as always delivered a wonderful presentation. Bonnie's mentorship of the younger presenters at the Prato conference, and indeed at all conferences that she attended was very much appreciated as well. Bonnie and I became good friends and it was a delight to visit her and husband, Doug at their house in upstate Michigan where they both were most relaxed and in their element, especially in the kitchen! I was fortunate to visit Bonnie and Doug last August when Bonnie was battling her cancer but we both had fond memories to reminisce on, of times past at scientific meetings, and in particular, the social engagements and meeting of old friends that always enlivened the meetings.

Dear Bonnie – you are dearly missed by us all but your science lives on! Vale Bonnie!

Remembering Bonnie



In Memoriam: *Dr Margarete Heck*

JCB Journal of Cell Biology

IN MEMORIAM

Margarete Heck (1959–2023): Cell biologist, geneticist, and incandescent social spark

Neville Cobbe[1], Francesca Di Cara[2], Allan C. Spradling[3], and Sharron Vass[4]

J. Cell Biol. 2024 Vol. 223 No. 1 e202311145

Read here: https://doi.org/10.1083/jcb.202311145

2024 GRC Meeting Report

Gordon Research Conference
on Proteolytic Enzymes and their Inhibitors
Barga, Italy 9-14 June 2024

GRC chaired by Jeanne Hardy and Galia Blum
GRS chaired by Robin Krystufek and Nathan Leborgne

Made possible by the generous support from our sponsors!

Gordon Research Conferences — Frontiers of Science
UMass Amherst
The Protein Society
Cambridge ProteinWorks
Novo Nordisk Pharmatech A/S | novo nordisk
Werewolf Therapeutics
CZECHSCIENCE.CZ
IOCB TECH
BioVendor LM
witec
International Proteolysis Society
The Company of Biologists

2024 GRC Awards

Congratulations to Nicholas Young from UCSF for receiving the Best Presentation Prize at the GRS, sponsored by the IPS.

I am incredibly honored to have received the International Proteolysis Society (IPS) Best Presentation Award at the 2024 Proteolytic Enzymes and Their Inhibitors Gordon Research Seminar (GRS). I am a PhD Candidate in the Chemistry and Chemical Biology Program at the University of California, San Francisco (UCSF) and co-advised by Dr. Charles Craik and Dr. Margaux Pinney.

At GRS, I presented work from my thesis project focused on adapting an emerging microfluidic platform towards the quantitative characterization of protease libraries (~103) in high throughput. As new mutations of SARS-CoV-2 main protease (Mpro) appear in the clinic, we wondered how to keep up with the virus as it mutates while functionally understanding how each of these mutations impact the activity and resistance of the protease. By using High Throughout Microfluidic Enzyme Kinetics (HT-MEK), we are now able to recombinantly express, purify, and characterize ~1792 Mpro variants per device enabling us to functionally dissect how individual mutations impact activity and inhibition in terms of kinetic parameters and inhibition constants. We are excited to be submitting this story in the next few months.

My time at the 2023 IPS Meeting and 2024 GRS/GRC was incredibly rewarding and important towards my career development. At these meetings, I have been able to connect with many leaders in academia and industry who were incredibly open in sharing their career path and providing constructive feedback on my project. These connections have extended past the meetings as well, where I have been able to seek advice and mentorship as I take next steps in my career path. The meeting chairs have a focus on promoting trainee development by providing opportunities for trainees to give talks and posters, setting up small group networking sessions, and promoting new collaborations. The international protease community has been incredibly welcoming, and they are excited to see us trainees succeed and make the next advances in the field. I look forward to completing my thesis this year and continuing my involvement in IPS.

Congratulations to the recipients of the inaugural Heck-Sloane Fellowship Travel Awards, in memory of IPS members Bonnie Sloane and Margarete Heck:

Henrique Baeta
Laura Donzelli
Rawad Hanna
Aleksander Haack
Brianna Hurysz
Nathan Leborgne
Gozde Ozcelik
Irina Sagarbarria
Kornelia Steindel
Magdalena Wądrzyk

Congratulations to the Poster Prize Winners:

Olivia Rickman
Anna Maywar
Martyna Majchrzak
Henrique Baeta
Dipon Saha
Armando Strong
Nicholas Young

San Diego Protease Symposium

Guy and Scott celebrated 32 years of the Salvesen Lab!

2024 Pacific Coast Protease Workshop

20th Anniversay Edition

CONGRATULATIONS to Timothy Harris from the Trader Lab at UC Irving for receiving the coveted Half Moon Bay Award for best presentation.

In April, the 20th annual Pacific Coast Protease Workshop, affectionately known as PCP, was held in Borrego Springs, California. Organized by Matt Bogyo, Antoine Dufour, Olivier Julien, Anthony O'Donoghue, and Scott Snipas, the meeting was attended by 51 delegates. There were 31 outstanding presentations, including invited talks from 5 'outside experts' - Lakshmi Wijeyewickrema, Laura Edgington-Mitchell, Howard Fearnhead, Ed Sturrock, and Charles Craik.

PCP Fun

In memory of The Silver Mullet

Meeting Announcement

FEBS 2024 ADVANCED COURSE

Proteolysis: at the interface between health and disease

14-18 September 2024 | Bled, Slovenia

Dear colleagues and friends,

it is our pleasure to invite you to this year's **FEBS workshop 'Proteolysis: at the interface between health and disease'**.

This workshop aims at addressing current issues, progress, and expansion taking place in the field of proteases, protein inhibitors and their mechanisms of control under physiological and pathological conditions. Several sections will be devoted to proteases, protease inhibitors, regulation and proteolytic control with emphasis on their role in diseases, as well as on novel therapeutic approaches, and in vivo imaging and diagnostic strategies, which tackle protease activities. Additionally, several sections will be devoted to tutoring and mentoring activities, which are crucial for the information and knowledge exchange among senior scientists and early career researchers.

We would like to thank FEBS for making it possible for us to organise this scientific meeting. Thank you also to IUBMB, which supported the meeting.

We are looking forward to seeing you in the enchanting environment of Bled in September!

Boris Turk,
Chair of the organizing committee

Deadlines

Applications Opening
26 April 2024

YTF and IUBMB Transconinenta Grant Application
30 July 2024

YTF and IUBMB Transconinenta Grant Notification
15 August 2024

Abstracts Submission
15 August 2024

Applications Closing
31 August 2024

Closing times: 23:59 (UTC+01:0

Galia Blum
The Hebrew University of Jerusalem, Israel

Hans Brandstetter
Paris-Lodron University of Salzburg, Austria

Klaudia Brix
Jacobs University Bremen, Germany

Marcin Drag
Wroclaw University of Technology, Poland

Ruth Geiss-Friedlander
University of Freiburg, Germany

James A. Huntington
University of Cambridge, UK

Janko Kos
University of Ljubljana, Slovenia

Gilles Lalmanach
University of Tours, France

Mohamed Lamkanfi
Ghent University, Belgium

Seamus Martin
Trinity College Dublin, Ireland

Jan Potempa
Jagiellonian University, Poland

Irit Sagi
Weizmann Institute of Science, Israel

Christopher Scott
Queen's University Belfast, UK

Kvido Stříšovský
Institute of Organic Chemistry and Biochemistry of the CAS, Czech Republic

Boris Turk
Jozef Stefan Institute, Slovenia

Dušan Turk
Jozef Stefan Institute, Slovenia

Olga Vasiljeva
CytomX Therapeutics Inc., USA

Silja Weßler
Paris-Lodron University Salzburg, Austria

Travel Awards for PhD Students and Postdocs are Available!!

For more information: https://proteolysis2024.febsevents.org/
Contact Boris: boris.turk@ijs.si

Meeting Announcement

Gordon Research Conferences

frontiers of science

Announcing the 2025 Gordon Research Conference on:

Plant Proteolysis

Integration and Regulation of Plant Proteolytic Pathways

Date and Location:
January 19 - 24, 2025
Renaissance Tuscany Il Ciocco
Lucca (Barga), Lucca, Italy

Organizers:
Chairs: Andreas Schaller & Marisa Otegui
Vice Chairs: Libo Shan & Byung-Ho Kang

Meeting Description:

The Plant Proteolysis field has grown rapidly in recent years, and tremendous progress has been made in many of its subareas. To further advance the field, an integrated view and interdisciplinary approaches to Plant Proteolysis are required. The 2025 GRC will thus join researchers studying basic, technical and applied aspects of Plant Proteolysis and covering a wide range of disciplines, including genetics, cell and molecular biology, chemistry and biochemistry, and systems biology. The 2025 GRC program includes invited experts in these areas and many talks selected from the abstracts. If you would like to be considered for an oral presentation, please apply before October 15, 2024.

Associated Gordon Research Seminar (GRS):
Proteolysis During Plant Development and Stress
January 18 - 19, 2025, Renaissance Tuscany Il Ciocco
chaired by Ariadna Gonzalez-Solis & Fausto Andres Ortiz Morea

More details and online application are available at:

https://www.grc.org/plant-proteolysis-conference/2025/

Exciting news for the protease field

Results from the Phase 3 ASPEN study validate cathepsin C as new therapeutic target with potential to address range of neutrophil serine protease-mediated diseases

« Our original innovative initiative is dedicated to establishing an efficient anti-proteolytic therapy upstream of pro-inflammatory neutrophil serine proteases by blocking their activating enzyme, cathepsin C ».

Cathepsin C, also called dipeptidyl peptidase 1 (DPP1), attracts more and more attention from both scientists and clinicians because of its role in the activation of neutrophil serine proteases (NSPs; elastase, proteinase 3 and cathepsin G) implicated in chronic inflammatory and auto-immune diseases. Promising preclinical and clinical data suggest that blockade of NSPs might ameliorate these conditions. Targeting NSPs by pharmacological inhibitors may appear as a simple, easy to manage approach. However, direct inhibition of NSPs has faced unresolved difficulties regarding the choice of protease to target or due to physicochemical properties of the inhibitors, prompting proposals for alternative approaches.

Patients with Papillon-Lefèvre syndrome have a genetically determined deficiency in cathepsin C but, reassuringly, do not exhibit marked immunodeficiency despite the absence of NSPs in immune defense cells. This observation has led to the conclusion that the pharmacological control of cathepsin C activity in bone marrow precursor cells could represent an attractive therapeutic strategy for NSP-mediated disorders including chronic obstructive pulmonary disease (COPD), alpha-1 antitrypsin deficiency, bronchiectasis, cystic fibrosis, pulmonary arterial hypertension, chronic rhinosinusitis ANCA-associated vasculitis, inflammatory bowel diseases and rheumatoid arthritis. Chronic inflammatory respiratory diseases affect over 1 billion people worldwide and cause the death of 4 million people every year. A further increase in the number of deaths from lung diseases is predicted until 2030, in particular from COPD.

A variety of cathepsin C inhibitors, developed by pharmaceutical companies and academic investigators, are currently being employed and evaluated in preclinical/clinical trials as anti-inflammatory drugs. A review of the therapeutic targeting of cathepsin C resulted from the first International Symposium on Cathepsin C, ISyCatC (Tours/France, April 2017) and was published in the journal Pharmacology and Therapeutics 1. This symposium launched the International Cathepsin C Consortium (ICat-CC) that we set up in 2016 thanks to the participation of world leading specialists on cathepsin C and its target serine proteases from academic labs and industry. ICat-CC is an international innovative consortium employing basic/translational and clinical research to establish proof of concepts for the repositioning of medicinal products blocking neutrophil serine protease activities.

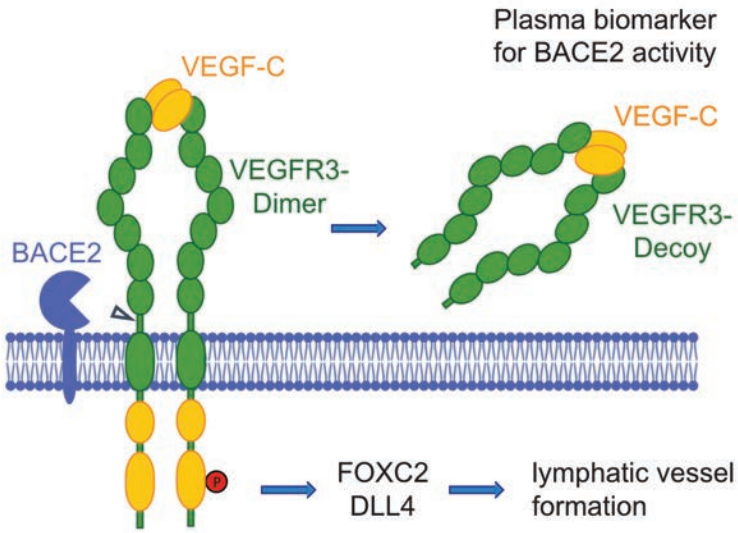
Cathepsin C inhibitors, brensocatib (Insmed Incorporated, NJ/USA), BI-1291583 (Boehringer Ingelheim, Germany) and HSK31858 (Chiesi Group (Italy) and Haisco Pharmaceutical (China)) are being evaluated in clinical trials in patients with chronic inflammatory lung diseases. Positive topline results from Phase 3 ASPEN study of brensocatib in patients with bronchiectasis were recently announced. If approved, brensocatib would be the first approved treatment for patients with bronchiectasis as well as the first approved cathepsin C inhibitor. Due to overlapping phenotypes and similar underpinning molecular mechanisms for a number of diseases associated with inflammation, positive effect in bronchiectasis patients could be translated directly to the potential treatment of other NSP-mediated inflammatory diseases. It is gratifying to see that the hard work of all colleagues from academic labs and industry and advocacy in cathepsin C field may have a clinical benefit.

1. Korkmaz, B.; Caughey, G. H.; Chapple, I.; Gauthier, F.; Hirschfeld, J.; Jenne, D. E.; Kettritz, R.; Lalmanach, G.; Lamort, A. S.; Lauritzen, C.; Legowska, M.; Lesner, A.; Marchand-Adam, S.; McKaig, S. J.; Moss, C.; Pedersen, J.; Roberts, H.; Schreiber, A.; Seren, S.; Thakker, N. S., Therapeutic targeting of cathepsin C: from pathophysiology to treatment. Pharmacol Ther 2018, 190, 202-236.

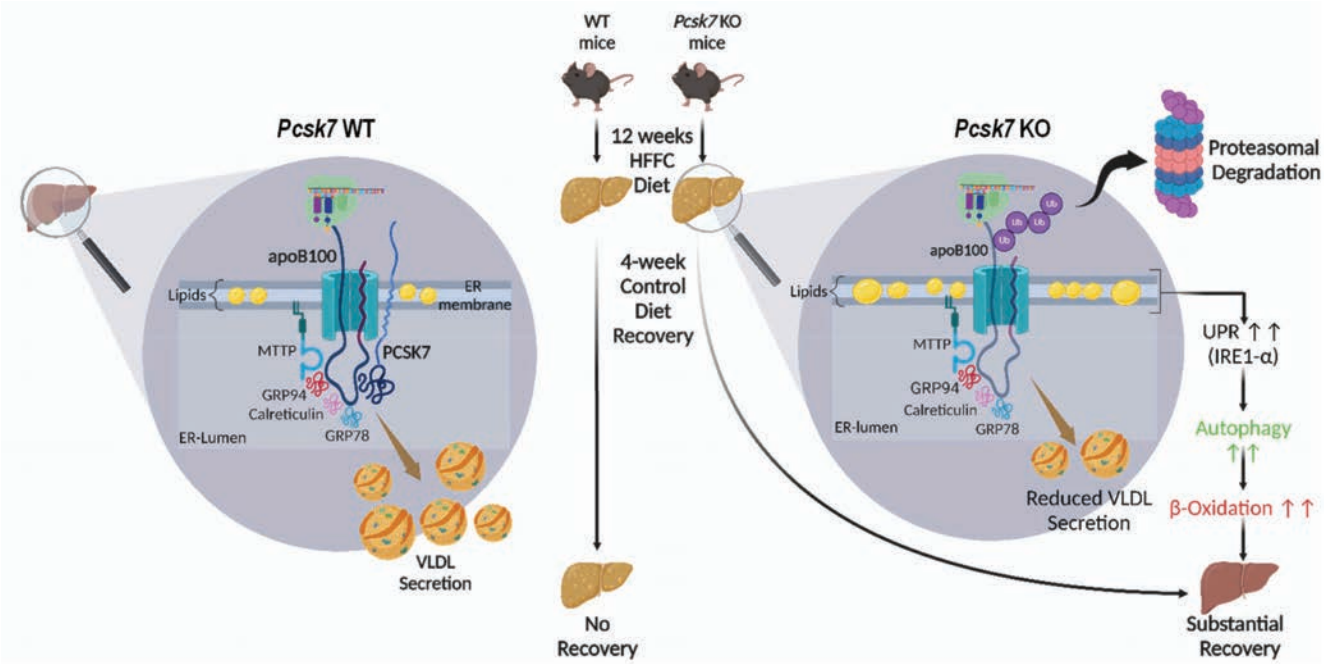
Brice KORKMAZ
INSERM UMR-1100
"Research Center for Respiratory Diseases (CEPR)" and Université de Tours
Tours, France

Recent Protease Papers

Schmidt A, Hrupka B, van Bebber F, Sunil Kumar S, Feng X, Tschirner SK, Aßfalg M, Müller SA, Hilger LS, Hofmann LI, Pigoni M, Jocher G, Voytyuk I, Self EL, Ito M, Hyakkoku K, Yoshimura A, Horiguchi N, Feederle R, De Strooper B, Schulte-Merker S, Lammert E, Moechars D, Schmid B, **Lichtenthaler SF.** (2024). **The Alzheimer's disease-linked protease BACE2 cleaves VEGFR3 and modulates its signaling.** J Clin Invest. e170550. doi: 10.1172/JCI170550. Epub ahead of print. PMID: 38888964.

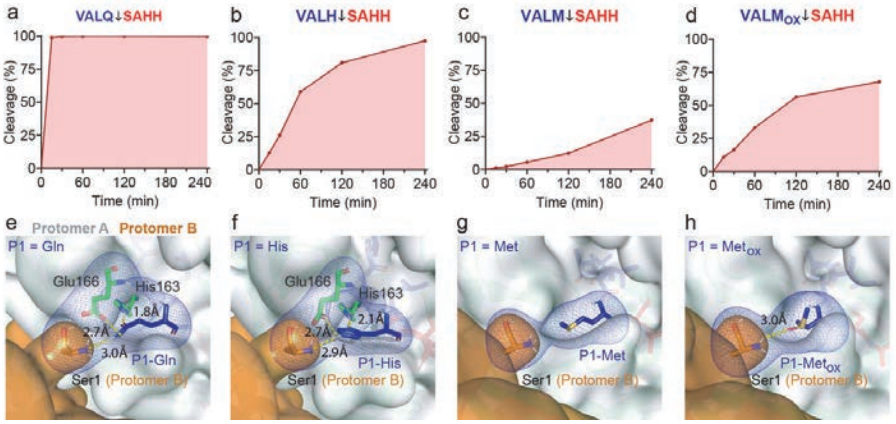


Sachan V, Le Dévéhat M, Roubtsova A, Essalmani R, Laurendeau JF, Garçon D, Susan-Resiga D, Duval S, Mikaeeli S, Hamelin J, Evagelidis A, Chong M, Paré G, Chernetsova E, Gao ZH, Robillard I, Ruiz M, Trinh VQ, Estall JL, Faraj M, Austin RC, Sauvageau M, Prat A, Kiss RS, **Seidah NG.** (2024). **PCSK7: A novel regulator of apolipoprotein B and a potential target against non-alcoholic fatty liver disease.** Metabolism. 150:155736. doi: 10.1016/j.metabol.2023.155736. PMID: 37967646.



Recent Protease Papers

Cesar Ramos de Jesus, H., Solis, N., Machado, Y., Pablos, I.M., Bell, P.A., Kappelhoff, R., Grin, P.M., Sorgi, C.A., Butler, G.S., and **Overall, C.M.** (2024) **Optimized Quenched Fluorescent Peptide Substrates of SARS-CoV-2 3CLpro Main Protease from Proteomic Identification of P6—P6' Active Site Specificity.** Journal of Virology 98, DOI. 10.1128/jvi.00049-24



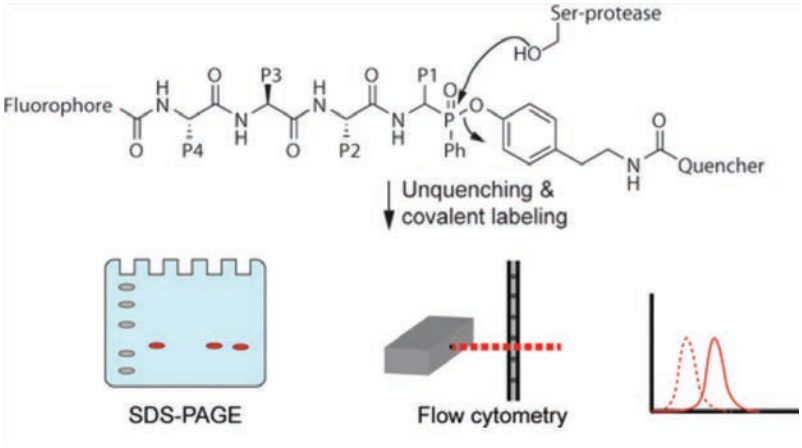
We determined the P6–P6' specificity for 3CLpro from >800 cleavage sites that we identified using 'Proteomic Identification of Cleavage site Specificity' (PICS). We characterized the P6–P6' active site specificity of SARS-CoV-2 3CLpro using proteome-derived peptide library screens, molecular modelling simulations and focussed positional peptide libraries. Cleavage occurred after the canonical P1-Gln and non-canonical P1-His and Met residues. In P1' we show that alanine and serine are cleaved 3x faster than glycine and the hydrophobic small amino acids Leu, Ile, or Val prevent cleavage of otherwise optimal non-prime sequences. A highly plastic S3' subsite accommodates P3'-His that displays stabilizing backbone h-bonds with Thr25 lying central in a 'threonine trio' (Thr24-Thr25-Thr26) in the P'-binding domain I. In characterizing non-canonical non-prime P1 specificity, we explored the unusual P1-Met specificity, discovering enhanced cleavage when in the oxidized state (P1-MetOX). We unveiled unexpected amino acid cooperativity at P1-Met with P3'-His and noncanonical P1-His with P2-Phe, and the importance of the threonine trio (Thr24-Thr25-Thr26) in the prime side binding domain I in defining prime side binding in SARS-CoV-2. Molecular docking simulations unveiled structure-activity relationships impacting 3CLpro-substrate interactions, and the role of these structural determinants was confirmed by MALDI-TOF-MS cleavage assays of P1'- and P3'-positional scanning peptide libraries carrying a 2nd optimal cut-site as an internal positive control. These data informed the design of two new and highly soluble 3CLpro quenched-fluorescent peptide substrates for improved FRET monitoring of 3CLproactivity with 15x improved sensitivity over current assays.

Vlok, M., Solis, N., Sadasivan, J., Mohamud, Y., Warsaba, R., Kizhakkedathu, J., Luo, H., **Overall, C.M.^ and Jan, E^.** (2024) **Identification of the Proteolytic Signature in CVB3-infected Cells.** Journal of Virology 98, JVI00498-24.

Coxsackievirus B3 (CVB3) encodes proteinases that are essential for processing of the translated viral polyprotein. Viral proteinases also target host proteins to manipulate cellular processes and evade innate antiviral responses to promote replication and infection. While some host protein substrates of the CVB3 3C and 2A cysteine proteinases have been identified, the full repertoire of targets is not known. Here, we utilize an unbiased quantitative proteomics-based approach termed terminal amine isotopic labeling of substrates (TAILS) to conduct a global analysis of CVB3 protease-generated N-terminal peptides in both human HeLa and mouse cardiomyocyte (HL-1) cell lines infected with CVB3. We identified >800 proteins that are cleaved in CVB3-infected HeLa and HL-1 cells including the viral polyprotein, known substrates of viral 3C proteinase such as PABP, DDX58 and HNRNPs M, K and D and novel cellular proteins. Network and GO-term analysis showed an enrichment in biological processes including immune response and activation, RNA processing and lipid metabolism. We validated a subset of candidate substrates that are cleaved under CVB3 infection and some are direct targets of 3C proteinase in vitro. Moreover, depletion of a subset of TAILS-identified target proteins decreased viral yield. Characterization of two target proteins showed that expression of 3Cpro-targeted cleaved fragments of emerin (EMD) and aminoacyl tRNA synthetase complex interacting multifunctional protein 2 (AIMP2) modulated autophagy and the NFkb pathway, respectively. The comprehensive identification of host proteins targeted during virus infection provides insights into the cellular pathways manipulated to facilitate infection.

Recent Protease Papers

Kahler, J.P.; Ji, S.; Speelman-Rooms, F.; Vanhoutte, R.; **Verhelst, S.H.L.** (2024) **Phosphinate Esters as Novel Warheads for Quenched Activity-Based Probes Targeting Serine Proteases.** ACS Chem Biol., in press. https://pubmed.ncbi.nlm.nih.gov/38913607/

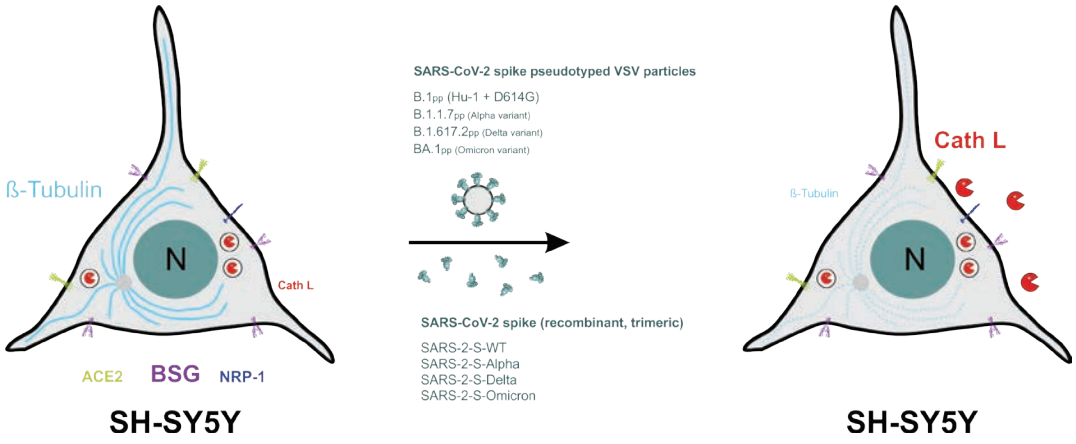


Skorenski, M.; Ji, S.; **Verhelst, S.H.L.** (2024) **Covalent activity-based probes for imaging of serine proteases.** Biochem. Soc. Trans., 24: 923-935. https://pubmed.ncbi.nlm.nih.gov/38629725/

Coene, J.; Wilms, S.; **Verhelst, S.H.L.** (2024) **Photoactivatable protease inhibitors: current status and perspectives, Chem. Eur. J., 30: e202303999.** https://pubmed.ncbi.nlm.nih.gov/38224181/

Zolg S, Donzelli L, **Geiss-Friedlander R.** (2024) **N-terminal processing by dipeptidyl peptidase 9: Cut and Go!** Biochimie. S0300-9084(24)00052-X. doi: 10.1016/j.biochi.2024.03.002. Epub ahead of print.

Oliveira BR, Nehlmeier I, Kempf AM, Venugopalan V, Rehders M, Ceniza MEP, Cavalcanti PATPV, Hoffmann M, Pöhlmann S, **Brix K.** (2024) **Cytoskeletal β-tubulin and cysteine cathepsin L deregulation by SARS-CoV-2 spike protein interaction with the neuronal model cell line SH-SY5Y.** Biochimie. S0300-9084(24)00044-0. doi: 10.1016/j.biochi.2024.02.006. Epub ahead of print. PMID: 38432290.

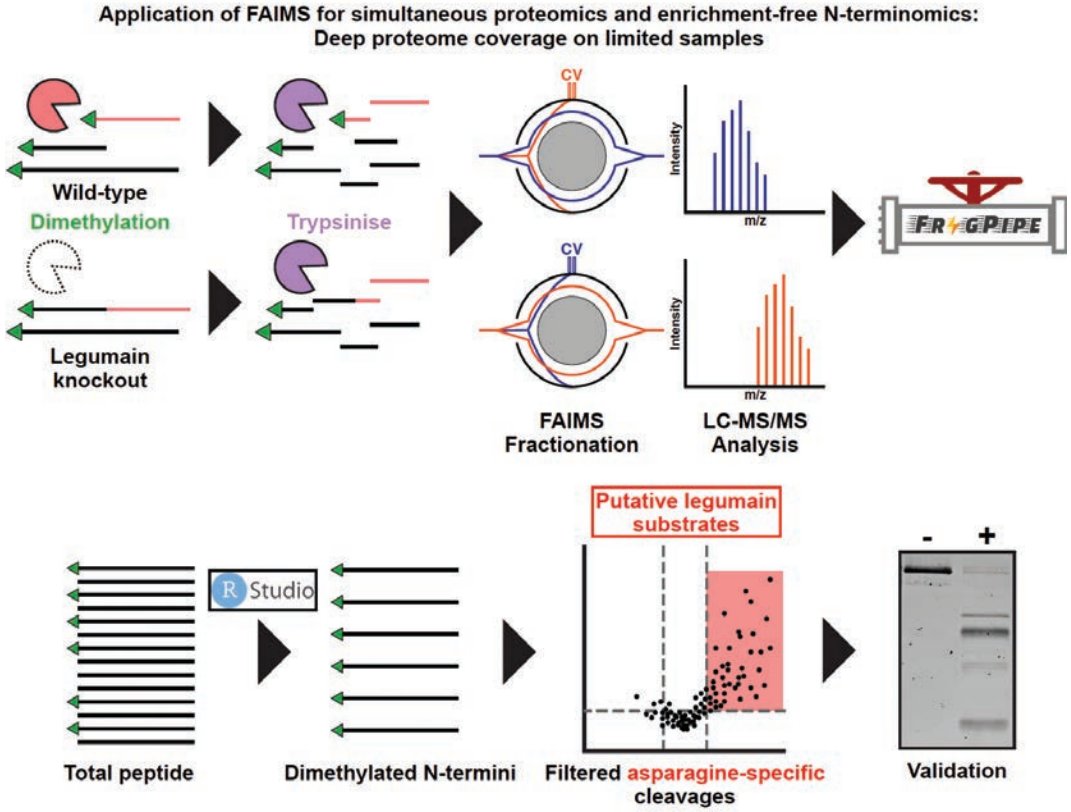


Recent Protease Papers

Radisky, E.S. (2024) **Extracellular proteolysis in cancer: Proteases, substrates, and mechanisms in tumor progression and metastasis.** J Biol Chem. 300(6):107347. doi: 10.1016/j.jbc.2024.107347. Epub 2024 May 6. PMID: 38718867; PMCID: PMC11170211.

Shoari, A., Khalili-Tanha, G., Coban, M.A., **Radisky, E.S.** (2024) **Structure and computation-guided yeast surface display for the evolution of TIMP-based matrix metalloproteinase inhibitors.** Front Mol Biosci. 10:1321956. doi: 10.3389/fmolb.2023.1321956. PMID: 38074088; PMCID: PMC10702220.

Ziegler AR, Dufour A, Scott NE, **Edgington-Mitchell LE.** (2024) **Ion Mobility-Based Enrichment-Free N-Terminomics Analysis Reveals Novel Legumain Substrates in Murine Spleen.** Mol Cell Proteomics. 23(2):100714. doi: 10.1016/j.mcpro.2024.100714.



Xu B, Anderson BM, Mountford SJ, Thompson PE, Mintern JD, **Edgington-Mitchell LE.** (2024). **Cathepsin X deficiency alters the processing and localisation of cathepsin L and impairs cleavage of a nuclear cathepsin L substrate.** Biol Chem. 405(5):351-365. doi: 10.1515/hsz-2023-0355. PMID: 38410910.

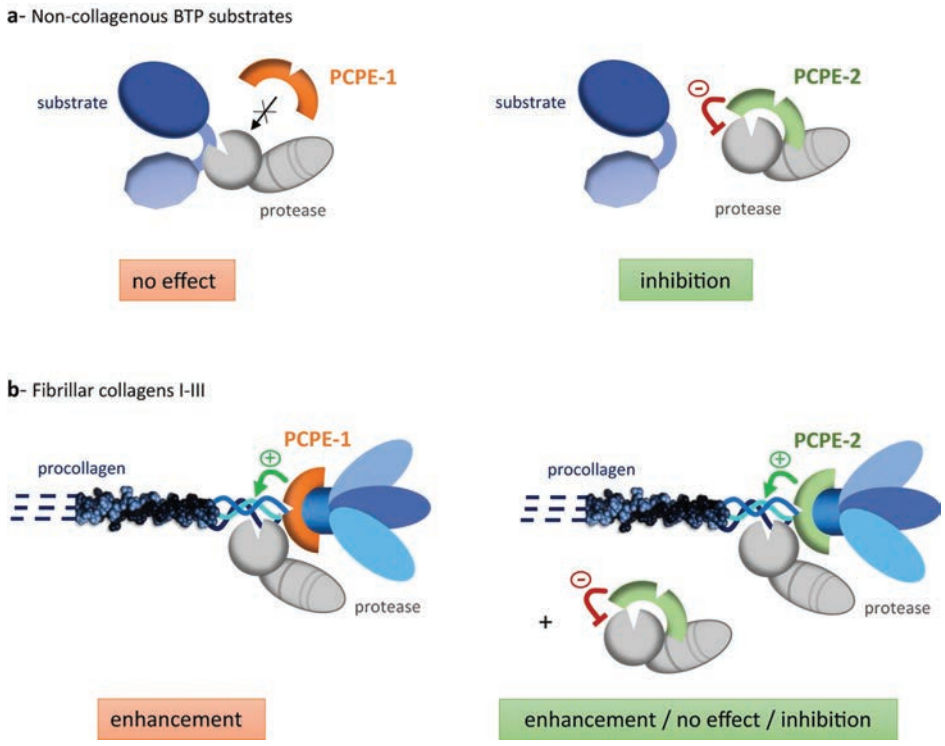
Xu B, Anderson BM, Mintern, JD, **Edgington-Mitchell LE**. (2024). **TLR9-dependent dendritic cell maturation promotes IL-6-mediated upregulation of cathepsin X**. Immunol Cell Biol. Epub ahead of print. http://doi.org/10.1111/imcb.12806

Recent Protease Papers

Hua Z, Watanabe R, Fukunaga T, Matsui Y, Matsuoka M, Yamaguchi S, Tanabe S, Yamamoto M, Tamura-Kawakami K, Takagi J, Kajita M, Futai E, **Shirakabe K.** (2024) **C-terminal amino acids in the type I transmembrane domain of L-type lectin VIP36 affect gamma-secretase susceptibility.** Biochem Biophys Res Commun. 696: 149504. doi: 10.1016/j.bbrc.2024.149504.

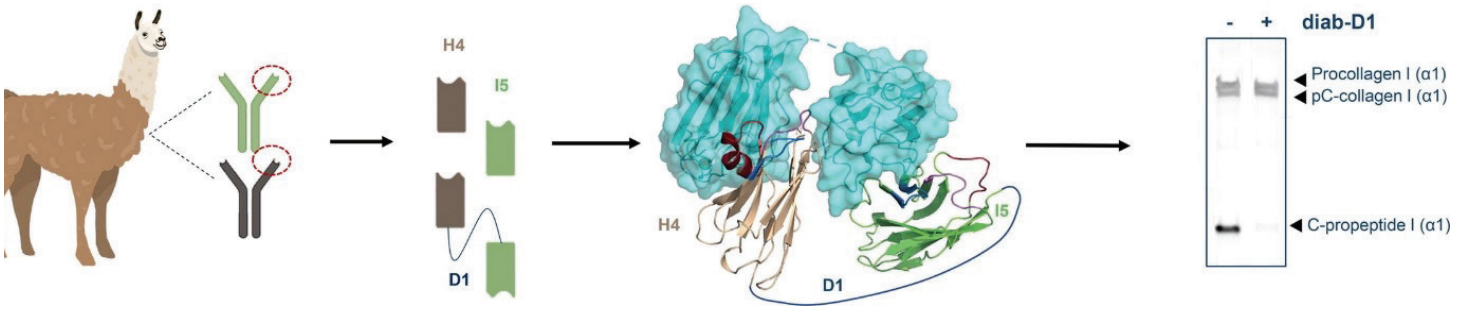
Vadon-Le Goff S, Tessier A, Napoli M, Dieryckx C, Bauer J, Dussoyer M, Lagoutte P, Peyronnel C, Essayan L, Kleiser S, Tueni N, Bettler E, Mariano N, Errazuriz-Cerda E, Fruchart Gaillard C, Ruggiero F, Becker-Pauly C, Allain JM, Bruckner-Tuderman L, Nyström A, **Moali C.** (2023) **Identification of PCPE-2 as the endogenous specific inhibitor of human BMP-1/tolloid-like proteinases.** Nat Commun. 14(1):8020. doi: 10.1038/s41467-023-43401-0. PMID: 38049428; PMCID: PMC10696041.

BMP-1/tolloid-like proteinases (BTPs) are major players in tissue morphogenesis, growth and repair. They act by promoting the deposition of structural extracellular matrix proteins and by controlling the activity of matricellular proteins and TGF-β superfamily growth factors. They have also been implicated in several pathological conditions such as fibrosis, cancer, metabolic disorders and bone diseases. Despite this broad range of pathophysiological functions, the putative existence of a specific endogenous inhibitor capable of controlling their activities could never be confirmed. Here, we show that procollagen C-proteinase enhancer-2 (PCPE-2), a protein previously reported to bind fibrillar collagens and to promote their BTP-dependent maturation, is primarily a potent and specific inhibitor of BTPs which can counteract their proteolytic activities through direct binding. PCPE-2 therefore differs from the cognate PCPE-1 protein and extends the possibilities to fine-tune BTP activities, both in physiological conditions and in therapeutic settings.



Recent Protease Papers

Lagoutte P, Bourhis JM, Mariano N, Gueguen-Chaignon V, Vandroux D, Moali C, **Vadon-Le Goff S.** (2024) **Mono- and Bi-specific Nanobodies Targeting the CUB Domains of PCPE-1 Reduce the Proteolytic Processing of Fibrillar Procollagens.** J Mol Biol. 436(16):168667. doi: 10.1016/j.jmb.2024.168667. Epub ahead of print. PMID: 38901640.



Yamano K, Sawada M, Kikuchi R, Nagataki K, Kojima W, Endo R, Kinefuchi H, Sugihara A, Fujino T, Watanabe A, Tanaka K, Hayashi G, Murakami H, **Matsuda N**. (2024) **Optineurin provides a mitophagy contact site for TBK activation**. EMBO J. 43: 754-779. doi: 10.1038/s44318-024-00036-1

Endo A, Fukushima T, Takahashi C, Tsuchiya H, Ohtake F, Ono S, Ly T, Yoshida Y, Tanaka K, Saeki Y, **Komada M.** (2024) **USP8 prevents aberrant NF-kB and Nrf2 activation by counteracting ubiquitin signals from endosomes.** J Cell Biol. 223(3): e202306013. doi: 10.1083/jcb.202306013