

PERSONAL DETAILS

Family name, First name: ACQUASALIENTE, LAURA

Researcher unique identifiers: Scopus ID: 56725712200; Researcher ID: AAE-2793-2021; ORCID: 0000-0001-6495-087

Nationality: Italian

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• EDUCATION AND KEY QUALIFICATIONS

March 2014 PhD in Molecular Sciences, XXVI Cycle

Dep. of Pharmaceutical and Pharmacological Sciences, University of Padova, Italy

Thesis Title: "Coagulation protein factors: discovering novel interactions of thrombin"

PhD Supervisor: Prof. V. De Filippis

March 2010 Master's degree cum laude in Chemistry and Pharmaceutical Technology

Dep. of Pharmaceutical and Pharmacological Sciences, University of Padova, Italy

Thesis Title: "New peptide inhibitors of thrombin from haematophagus parasites: chemical synthesis and characterization of full-length haemadin (1-57) from *Haemadipsa sylvestris* and mini-haemadin (1-10)"

Thesis Supervisor: Prof. V. De Filippis

• CURRENT POSITION

2023 - 2026 Tenure track researcher (RTDB) in Biochemistry

Dep. of Pharmaceutical and Pharmacological Sciences, University of Padova, Italy

• PREVIOUS POSITION

2019 – 2022 Tenure track researcher (RTDA) in Biochemistry

Dep. of Pharmaceutical and Pharmacological Sciences, University of Padova, Italy

• FELLOWSHIPS

2017 – 2019 Postdoctoral Fellow

Dep. of Biochemistry and Molecular Biology, Saint Louis University, MO-USA

Supervisor Prof. E. Di Cera

NIH Grant 2017-2019: Structural determinants of prothrombin activation

2014 – 2016 Postdoctoral Fellow

Chemical Department, University of Padova, Italy

Supervisor Proff. A. Polimeno and V De Filippis

Excellence Research Project CaRiPaRo-2011: Modeling and monitoring motions in proteins: cardiovascular disease at the molecular level

Career breaks, diverse career paths and major life events

4th October 2020 – 4th March 2021: Career break of 5 months for maternity leave

(Executive Decree n. 3673 4/November/2020)

27th August 2023 – 26th January 2024: Career break of 5 months for maternity leave

(Executive Decree n. 3441 5/September/2023)

RESEARCH ACHIEVEMENTS AND PEER RECOGNITION

Research achievements

Over the last decade, my scientific interest has been focused on elucidating non-canonical mechanisms linking protein misfolding, amyloid formation, and dysregulation of the coagulation cascade. Using a combination of advanced methodologies—including mass spectrometry (LC-MS/MS for enzyme specificity profiling, HDX-MS for conformational dynamics), biophysical and spectroscopic approaches (SPR, fluorescence, circular dichroism, dynamic light scattering), and functional assays of hemostasis—we have demonstrated that transthyretin (TTR) amyloid fibrils can act as polymeric scaffolds for the activation of factor XII and the contact system, thereby promoting pathological coagulation (paper to be submitted). Building on this expertise, we are now particularly interested in investigating amyloidogenic mutations of TTR, which provide an ideal framework to

dissect how specific structural alterations drive aggregation, modulate molecular interactions, and contribute to thrombosis.

In parallel, we are studying how natural compounds and small molecules can modulate the aggregation of α -synuclein, a protein implicated in neurodegenerative disorders. By combining biophysical characterization, aggregation assays, and molecular interaction studies, we aim to identify chemical modulators capable of altering amyloid formation, providing insights into potential therapeutic strategies for protein aggregation-related pathologies.

These studies collectively allow us to define novel biochemical mechanisms of amyloidosis-associated coagulopathies and protein aggregation, while also exploring avenues for therapeutic intervention.

List of research outputs

1. Pagotto A., Uliana F., Cavedon E., Nordio G., Pierangelini A., **Acquasaliente L.**, Macchia M.L., Bellanda M., Gatto B., De Silvestro G., Marson P., Gregori D., Simioni P., Picotti P., De Filippis V. The main protease (M^{pro}) from SARS-CoV-2 triggers plasma clotting in vitro by activating coagulation factors VII and FXII. *Commun Biol.* **2025** Aug 1;8(1):1145. doi: 10.1038/s42003-025-08570-2.
2. Rizzotto E., Pierangelini A., Fongaro B., Leri M., Inciardi I., Trolese P., De Filippis V., Bucciantini M., **Acquasaliente L.**[#], Polverino de Laureto P. DOPAC as a modulator of α -Synuclein and E46K interactions with membrane: Insights into binding dynamics. *Int J Biol Macromol.* **2025** ;294:139427. doi: 10.1016/j.ijbiomac.2024.139427.
3. Coluccino G., Negro A., Filippi A., Bean C., Muraca V.P., Gissi C., Canetti D., Mimmi M.C., Zamprogno E., Ciscato F., **Acquasaliente L.**, De Filippis V., Comelli M., Carraro M., Rasola A., Gerle C., Bernardi P., Corazza A., Lippe G. N-terminal cleavage of cyclophilin D boosts its ability to bind F-ATP synthase. *Commun Biol.* **2024**;7(1):1486. doi: 10.1038/s42003-024-07172-8.
4. De Filippis V., **Acquasaliente L.**, Pierangelini A., Marin O. Chemical Synthesis and Structure-Activity Relationship Studies of the Coagulation Factor Xa Inhibitor Tick Anticoagulant Peptide from the Hematophagous Parasite *Ornithodoros moubata*. *Biomimetics (Basel).* **2024**;9(8):485. doi: 10.3390/biomimetics9080485.
5. **Acquasaliente L.**[^], Pierangelini A., Pagotto A., Pozzi N., De Filippis V. From haemadin to haemanorm: Synthesis and characterization of full-length haemadin from the leech *Haemadipsa sylvestris* and of a novel bivalent, highly potent thrombin inhibitor (haemanorm). *Protein Sci.* **2023** Dec;32(12):e4825. doi: 10.1002/pro.4825.
6. Mazzocato Y., Perin S., Morales-Sanfrutos J., Romanyuk Z., Pluda S., **Acquasaliente L.**, Borsato G., De Filippis V., Scarso A., Angelini A. A novel genetically-encoded bicyclic peptide inhibitor of human urokinase-type plasminogen activator with better cross-reactivity toward the murine orthologue. *Bioorg Med Chem.* **2023** Nov 15;95:117499. doi: 10.1016/j.bmc.2023.117499.
7. Pavan M., Fanti C.D., Lucia A.D., Canato E., **Acquasaliente L.**, Sonvico F., Delgado J., Hicks A., Torrelles J.B., Kulkarni V., Dwivedi V., Zanellato A.M., Galesso D., Pasut G., Buttini F., Martinez-Sobrido L., Guarise C. Aerosolized sulfated hyaluronan derivatives prolong the survival of K18 ACE2 mice infected with a lethal dose of SARS-CoV-2. *European Journal of Pharmaceutical Sciences* **2023**, 187:106489. doi: 10.1016/j.ejps.2023.106489.
8. Guarise C., **Acquasaliente L.**, Pasut G., Pavan M., Soato M., Garofolin G., Beninatto R., Giacomel E., Sartori E., Galesso D. The role of high molecular weight hyaluronic acid in mucoadhesion on an ocular surface model. *Journal of the Mechanical Behavior of Biomedical Materials* **2023**, 105908, doi:10.1016/j.jmbbm.2023.105908.
9. **Acquasaliente L.**^{^#}, De Filippis V. The Role of Proteolysis in Amyloidosis. *International Journal of Molecular Sciences* **2023**, 24, 699. doi: 10.3390/ijms24010699
10. Di Liddo R., Verona M., Vaccarin C., **Acquasaliente L.**, Schrenk S., Piccione M., Cenzi C., De Franco M., Dal Prà M., Ribaudo R., Ferlin M.G., Conconi M.T., Chilin A., Gandin V., Marzaro G. Preliminary discovery of Small Molecule Inhibitors of Epidermal Growth Factor Receptor (EGFR) that bind to the extracellular domain. *Cancer* **2022**, <https://doi.org/10.3390/cancers14153647>
11. **Acquasaliente L.**[^], Pontarollo G.[^], Radu C.M., Peterle D., Artusi I., Pagotto A., Uliana F., Negro A., Simioni P., De Filippis V. Exogenous human α -synuclein acts in vitro as a mild platelet

antiaggregant inhibiting α -thrombin-induced platelet activation. *Scientific Reports* **2022**, 12, 9880. doi.org/10.1038/s41598-022-12886-y.

12. Gasparotto M., Hernandez Gomez Y.S., Peterle D., Grinzato A., Zen F., Pontarollo G., **Acquasaliente L.**, Scapin G., Bergantino E., De Filippis V., Filippini F. NOG-Derived Peptides Can Restore Neuritogenesis on a CRASH Syndrome Cell Model. *Biomedicines*. **2022**, 4;10(1):102, doi: 10.3390/biomedicines10010102.
13. Scapin G., Gasparotto M., Peterle D., Tescari S., Porcellato E., Piovesan A., Righetto I., **Acquasaliente L.**[#], De Filippis V., Filippini F. A conserved Neurite Outgrowth and Guidance motif with biomimetic potential in neuronal Cell Adhesion Molecules. *Computational and Structural Biotechnology Journal*. **2021**, 19:5622-5636, doi: 10.1016/j.csbj.2021.10.005.
14. Palazzi L.; Fongaro B.; Leri M.; **Acquasaliente L.**; Stefani M.; Bucciantini M.; Polverino de Laureto P. Structural Features and Toxicity of α -Synuclein Oligomers Grown in the Presence of DOPAC. *International Journal of Molecular Sciences* **2021**, 22, 6008, doi: 10.3390/ijms22116008.
15. **Acquasaliente L.**[^], Dal Lin C.[^], Iliceto S. De Filippis V.; Vitiello G.; Tona F. Von Willebrand Factor Multimers and the Relaxation Response: A One-Year Study. *Entropy* **2021**, 23, 447, doi: 10.3390/e23040447.
16. Ulian, F.[^]; Vizovišek M.[^]; **Acquasaliente L.**; Ciuffa R.; Fossati A.; Frommelt F.; Goetze S.; Wollscheid B.; Gstaiger, M.; De Filippis V.; Auf dem Keller U., Aebersold R. Mapping specificity, cleavage entropy, allosteric changes and substrates of blood proteases in a high-throughput screen. *Nature Communication* **2021**, 12, 1693, doi:10.1038/s41467-021-21754-8.
17. De Toni L.; Sabovic I.; De Filippis V.; **Acquasaliente L.**; Peterle D.; Guidolin D.; Sut S.; Di Nisio A.; Foresta C.; Garolla A. Sperm Cholesterol Content Modifies Sperm Function and TRPV1-Mediated Sperm Migration. *International Journal of Molecular Sciences* **2021**, 22, doi:10.3390/ijms22063126.
18. Di Nisio A.; Rocca M.S.; De Toni L.; Sabovic I.; Guidolin D.; Dall'Acqua S.; **Acquasaliente L.**; De Filippis V.; Plebani M.; Foresta C. Endocrine disruption of vitamin D activity by perfluorooctanoic acid (PFOA). *Scientific Reports* **2020**, 10, 16789, doi:10.1038/s41598-020-74026-8.
19. Di Nisio A.; Rocca M.S.; Sabovic I.; De Rocco Ponce M.; Corsini C.; Guidolin D.; Zanon C.; **Acquasaliente L.**; Carosso A.R.; De Toni L.; Foresta C. Perfluorooctanoic acid alters progesterone activity in human endometrial cells and induces reproductive alterations in young women. *Chemosphere* **2020**, 242, 125208, doi:10.1016/j.chemosphere.2019.125208.
20. **Acquasaliente L.**[^]; Pelc L.A.; Di Cera E. Probing prothrombin structure by limited proteolysis. *Scientific Reports* **2019**, 9, 6125, doi:10.1038/s41598-019-42524-z.
21. Ricatti J.; **Acquasaliente L.**; Ribaud G.; De Filippis V.; Bellini M.; Llovera R.E.; Barollo S.; Pezzani R.; Zagotto G.; Persaud K.C.; Mucignat-Caretta C. Effects of point mutations in the binding pocket of the mouse major urinary protein MUP20 on ligand affinity and specificity. *Scientific Reports* **2019**, 9, 300, doi:10.1038/s41598-018-36391-3.
22. Di Nisio A.; Sabovic I.; Valente U.; Tescari S.; Rocca M.S.; Guidolin D.; Dall'Acqua S.; **Acquasaliente L.**; Pozzi N.; Plebani M.; Garolla A.; Foresta C. Endocrine Disruption of Androgenic Activity by Perfluoroalkyl Substances: Clinical and Experimental Evidence. *The Journal of Clinical Endocrinology & Metabolism* **2019**, 104, 1259-1271, doi:10.1210/jc.2018-01855.
23. Chakraborty P.; **Acquasaliente L.**; Pelc L.A.; Di Cera E. Interplay between conformational selection and zymogen activation. *Scientific Reports* **2018**, 8, 4080, doi:10.1038/s41598-018-21728-9.
24. Pengo V.; Del Ross T.; Ruffatti A.; Bison E.; Cattini M.G.; Pontara E.; Testa S.; Legnani C.; Pozzi N.; Peterle D.; **Acquasaliente L.**; De Filippis V.; Denas G. Lupus anticoagulant identifies two distinct groups of patients with different antibody patterns. *Thrombosis Research* **2018**, 172, 172-178, doi:10.1016/j.thromres.2018.11.003.
25. De Filippis V., Pozzi N., **Acquasaliente L.**, Artusi I., Pontarollo G., Peterle D.. Protein engineering by chemical methods: Incorporation of nonnatural amino acids as a tool for studying protein folding, stability, and function. *Biotechnology and Applied Biochemistry* **2018**. doi.org/10.1002/pep2.24090.

26. De Filippis V., **Acquasaliente L.**, Pontarollo G., Peterle D.. Noncoded amino acids in protein engineering: Structure-activity relationship studies of hirudin-thrombin interaction. *Biotechnology and Applied Biochemistry* **2018**, 65(1) 69-80. doi: 10.1002/bab.1632.
27. Fagagnini A.; Pica A.; Fasoli S.; Montioli R.; Donadelli M.; Cordani M.; Butturini E.; **Acquasaliente L.**; Picone D.; Gotte G. Onconase dimerization through 3D domain swapping: structural investigations and increase in the apoptotic effect in cancer cells. *Biochemical Journal* **2017**, 474, 3767-3781, doi:10.1042/bcj20170541.
28. Galazzo L.; Maso L.; De Rosa E.; Bortolus M.; Doni D.; **Acquasaliente L.**; De Filippis V.; Costantini P.; Carbonera D. Identifying conformational changes with site-directed spin labeling reveals that the GTPase domain of HydF is a molecular switch. *Scientific Reports* **2017**, 7, 1714, doi:10.1038/s41598-017-01886-y.
29. Pontarollo G.[^]; **Acquasaliente L.[^]**; Peterle D.; Frasson R.; Artusi I.; De Filippis V. Non-canonical proteolytic activation of human prothrombin by subtilisin from *Bacillus subtilis* may shift the procoagulant-anticoagulant equilibrium toward thrombosis. *Journal of Biological Chemistry* **2017**, 292, 15161-15179, doi:10.1074/jbc.M117.795245.
30. **Acquasaliente L.**; Peterle D.; Tescari S.; Pozzi N.; Pengo V.; De Filippis V. Molecular mapping of alpha-thrombin (alphaT)/beta2-glycoprotein I (beta2Gpl) interaction reveals how beta2Gpl affects alphaT functions. *Biochemical Journal* **2016**, 473, 4629-4650, doi:10.1042/bcj20160603.
31. Pozzi N.; Zerbetto M.; **Acquasaliente L.**; Tescari S.; Frezzato D.; Polimeno A.; Gohara D.W.; Di Cera E.; De Filippis V. Loop Electrostatics Asymmetry Modulates the Preexisting Conformational Equilibrium in Thrombin. *Biochemistry* **2016**, 55, 3984-3994, doi:10.1021/acs.biochem.6b00385.
32. Vecellio Reane D.; Vallese F.; Checchetto V.; **Acquasaliente L.**; Butera G.; De Filippis V.; Szabo I.; Zanotti G.; Rizzuto R.; Raffaello A. A MICU1 Splice Variant Confers High Sensitivity to the Mitochondrial Ca(2+) Uptake Machinery of Skeletal Muscle. *Molecular Cell* **2016**, 64, 760-773, doi:10.1016/j.molcel.2016.10.001.
33. Sokolov A.V.[^]; **Acquasaliente L.[^]**; Kostevich V.A.; Frasson R.; Zakharova E.T.; Pontarollo G.; Vasilyev V.B.; De Filippis V. Thrombin inhibits the anti-myeloperoxidase and ferroxidase functions of ceruloplasmin: relevance in rheumatoid arthritis. *Free Radical Biology & Medicine* **2015**, 86, 279-294, doi:10.1016/j.freeradbiomed.2015.05.016.
34. Pozzi N.[^]; **Acquasaliente L.[^]**; Frasson R.; Cristiani A.; Moro S.; Banzato A.; Pengo V.; Scaglione G.L.; Arcovito A.; De Cristofaro R.; De Filippis V.. beta2 -Glycoprotein I binds to thrombin and selectively inhibits the enzyme procoagulant functions. *Journal of Thrombosis and Haemostasis* **2013**, 11, 1093-1102, doi:10.1111/jth.12238.
35. Banzato A.; Frasson R.; **Acquasaliente L.**; Bison E.; Bracco A.; Denas G.; Cuffaro S.; Hoxha A.; Ruffatti A.; Iliceto S.; De Filippis V., Pengo V. Circulating beta2 glycoprotein I-IgG anti-beta2 glycoprotein I immunocomplexes in patients with definite antiphospholipid syndrome. *Lupus* **2012**, 21, 784-786, doi:10.1177/0961203312440347.

[^] equal contribution

[#] corresponding author

Peer recognition

Fellowships, Awards, and Recognitions

- **2023:** Award for Poster Presentation at the “ECTH, European Congress on Thrombosis and Haemostasis” (Valencia, Spain). This recognition underscores the impact of my research on thrombosis and haemostasis.
- **2020:** Award for Poster Presentation at the “ECTH, European Congress on Thrombosis and Haemostasis” (Ghent, BE), acknowledging my ongoing contributions to the understanding of blood coagulation mechanisms.
- **2017:** Recognition for Scientific Contribution from the Editorial Board of the *Journal of Biological Chemistry* for the paper “Non-canonical proteolytic activation of human prothrombin by subtilisin

from *Bacillus subtilis* may shift the procoagulant-anticoagulant equilibrium toward thrombosis," selected among the 50 best publications of the year from over 6,600 articles.

- **2017-2019:** *NIH Postdoctoral Fellowship* on "Structural determinants of prothrombin activation" funded by Saint Louis University, USA, supporting my research on the molecular mechanisms of blood coagulation.
- **2016:** *PRAT Postdoctoral Fellowship* on "Chemical modifications of coagulation proteins in thrombotic diseases," funded by the University of Padova, advancing the understanding of protein modifications in thrombosis.
- **2015:** *HIC LABOR AWARD 2014* for Best PhD Thesis, awarded by the Accademia Olimpica (Vicenza, Italy).
- **2014-2015:** *CaRiPaRo Postdoctoral Fellowship* on "Modeling and Monitoring Motions in Proteins: Cardiovascular diseases at the molecular level," funded by the University of Padova, Italy.

Contributions to Scientific Conferences

- **2025:** Member of the committee for poster evaluation "Protein Misfolding and Aggregation in Disease" (Mantova, Italy)
- **2023:** *Speaker* at "Focus on Neuropharmacology and Brain Delivery" (Monash University + DSF, University of Padova). Presentation: "Catechols as a new therapeutic strategy against Parkinson's disease."
- **2022:** *Organizer and Chair* of the "Surface Plasmon Resonance (SPR) Biacore-T200 Demo On-site" workshop, University of Padova.
- **2021:** *Invited Speaker* at the "45th FEBS Virtual Congress" (Ljubljana, Slovenia). Presentation: "β2-Glycoprotein I (β2GPI) is a Fibrinogen Binding Protein."
- **2020-2021:** *Speaker* at the International Pharmaceutical Biotechnologies program (University of Padova). Presentation: "Protein-Protein Interaction: The application of Surface Plasmon Resonance Technique."
- **2018:** *Speaker* at the "Special Event of Biochemistry Department Retreat" (Danforth Plant Science Center, St. Louis, USA). Presentation: "Prothrombin Domains on the Mechanism of Activation."
- **2016:** *Speaker* at the "Special Event UNIPD and East China University" (University of Padova). Presentation: "Discovering Novel Interactions of Thrombin."
- **2013:** *Speaker (Selected)* at the "XXIV Congress of the International Society on Thrombosis and Haemostasis" (Amsterdam, NL). Presentation: "Alpha-synuclein binds to thrombin exosites and inhibits thrombin-mediated platelet aggregation."
- **2013:** *Invited Speaker* at the "V School of Vegetal Physiopathology" (Pieve di Tesino, Italy). Seminar: "Protein-Protein Interaction: Structure Biology and Experimental Methods."

ADDITIONAL INFORMATION

Technical expertise:

- Protein Chemistry Techniques: solid-phase peptide synthesis; chemical modification of proteins
- Analytical Techniques: protein purification; chromatographic and electrophoretic techniques; mass spectrometry
- Spectroscopic Techniques: UV-Vis absorption; circular dichroism; fluorescence spectroscopy; dynamic Light Scattering (DLS); basic Small-Angle X-ray scattering
- Molecular Recognition: Surface Plasmon Resonance (SPR); Isothermal Titration Calorimetry (ITC); Differential Scanning Calorimetry (DSC)
- Enzyme Kinetics: determination of enzyme kinetic constants under steady-state and rapid conditions (stopped flow); determination of enzyme inhibition constants
- Molecular Biology techniques: protein expression in mammalian cells, yeast and *E.coli*
- Practical Haemostasis Techniques: determination of clinical coagulation parameters (TT, PT, aPTT, SCT); fibrin generation; platelet agglutination; multimer size distribution of von Willebrand Factor by agarose gel-electrophoresis
- Bioinformatics: data analysis software (Origin 8.0, SigmaPlot); interrogation of databases for

protein identification and characterization (EXPASY, MASCOT); molecular visualization and modelling software (PyMol, HyperChem, Chimera, MOE); molecular docking software (HEX, ClusPro, HPEPdock); protein structure analysis software (BLUUES, APBS)

- Protein Structure Techniques: robot-automated crystallization; micro-scale crystallization techniques (hanging drop and sitting drop); basic knowledge of NMR; HDX mass spectrometry

OTHER ACTIVITIES

Editorial and Reviewing Activities

- Since 2024: Member of the Editorial Board as Associate Editor for Frontiers in Molecular Biosciences, section Molecular Diagnostics and Therapeutics.
- Since 2023: Member of the Editorial Board as Review Editor – Community Reviewer for Frontiers in Molecular Biosciences, section Structural Biology.
- Since 2023: Guest Editor for the Special Issue “Biomimetic Peptides and Proteins” in the international scientific journal Biomimetics (MDPI, ISSN 2313-7673). https://www.mdpi.com/journal/biomimetics/special_issues/LJF4D2CH39
- Since 2017: Regular reviewer for several international scientific journals, including Scientific Reports, European Journal of Pharmaceutical Sciences, Journal of Clinical Medicine, Cells, Biochemistry and Biophysics Reports, and International Journal of Molecular Sciences.

Affiliation to Scientific Societies

- Since 2022: Member of the Italian Society of Biochemistry and Molecular Biology (Società Italiana di Biochimica e Biologia Molecolare).
- Since 2020: Member of the International Society on Thrombosis and Haemostasis.
- 2017–2019: Member of the International Fibrinogen Research Society.