13TH INTERNATIONAL CONFERENCE ON HIGH PRESSURE
BIOSCIENCE AND BIOTECHNOLOGY
UNIVERSITY OF AVEIRO • 31st August - 3rd September 2026

13th International Conference on High Pressure Bioscience and Biotechnology (HPBB)

Conference Booklet

University of Aveiro

Aveiro, 31st August – 3rd September 2026

The International Association of High Pressure Bioscience and Biotechnology (HPBB)

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Welcome message

Dear colleagues,

It is a great pleasure to welcome you all to the 13th International Conference on High Pressure Bioscience and Biotechnology (HPBB) at the University of Aveiro, in the wonderful and picturesque city of Aveiro, Portugal, from 31st of August to 3rd of September 2026.

The HPBB conference has been a meeting point connecting researchers working across Food Science, Biosciences and Biotechnology related to High Pressure, united by a shared interest in the effects and applications of this increasingly interesting Food and Life variable. We hope that the 13th HPBB continues this tradition, providing an enrichment and friendly environment for the exchange of ideas to promote research and discussion of emerging topics and Biotechnological applications involving High Pressure and Food and Life.

The University of Aveiro is a young and dynamic public university with a strong international projection and a well-established tradition in High Pressure research, particularly in Food Science and Biotechnology. It is recognized for its interdisciplinary approach, close ties between fundamental science and applied research, commitment to innovation and knowledge transfer to the industry, making it stimulating and welcoming environment to host the 13th HPBB.

Aveiro, often referred to as the “Portuguese Venice”, is a coastal city known for its canals, traditional *Moliceiro* boats, and unique and distinctive *Art Nouveau* architecture. Beyond its scientific and academic environment, Aveiro region offers a strong connection to Portuguese culture, from its maritime heritage and cuisine to its welcoming atmosphere and pace of life, being the national centre of the famous and peculiar Portuguese codfish cuisine, supported by a dynamic industry in this field, having as pinnacle the yearly already internationally famous codfish festival taking place in August in Ílhavo, about 15 minutes away from the city of Aveiro. We hope all this will provide an inspiring set up for fruitful scientific discussions, as well as opportunities for informal interaction and cultural discovery.

On behalf of the Organizing and Scientific Committees, we warmly welcome you at the 13th HPBB, as we hope you have a stimulating and wonderful conference that reflects both the scientific excellence of the HPBB community and the hospitality of Portugal.

Sincerely yours, on behalf of the organizing committee,

Jorge A. Saraiva
Chair

Carlos A. Pinto
Co-Chair

Organizing Committee

	**Jorge Saraiva** *Chair*	Chemistry Department, University of Aveiro
	**Carlos Pinto** *Co-chair*	Chemistry Department, University of Aveiro
	**Alireza Ganjeh** *PhD Student*	Chemistry Department, University of Aveiro Faculty of Pharmacy, University of Porto
	**Nicole Moreira** *PhD Student*	Chemistry Department, University of Aveiro Colab4Food Faculty of Pharmacy, University of Porto
	**Ana Martins** *PhD Student*	Chemistry Department, University of Aveiro
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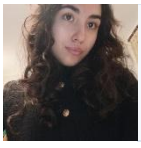
	**Guilherme Pereira** *MSc Student*	Chemistry Department, University of Aveiro
	**Carolina Antunes** *BSc Student*	Chemistry Department, University of Aveiro
	**Margarida Loureiro** *BSc Student*	Chemistry Department, University of Aveiro


BERICAP

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- **Abram Aertsen**, KU Leuven, Belgium
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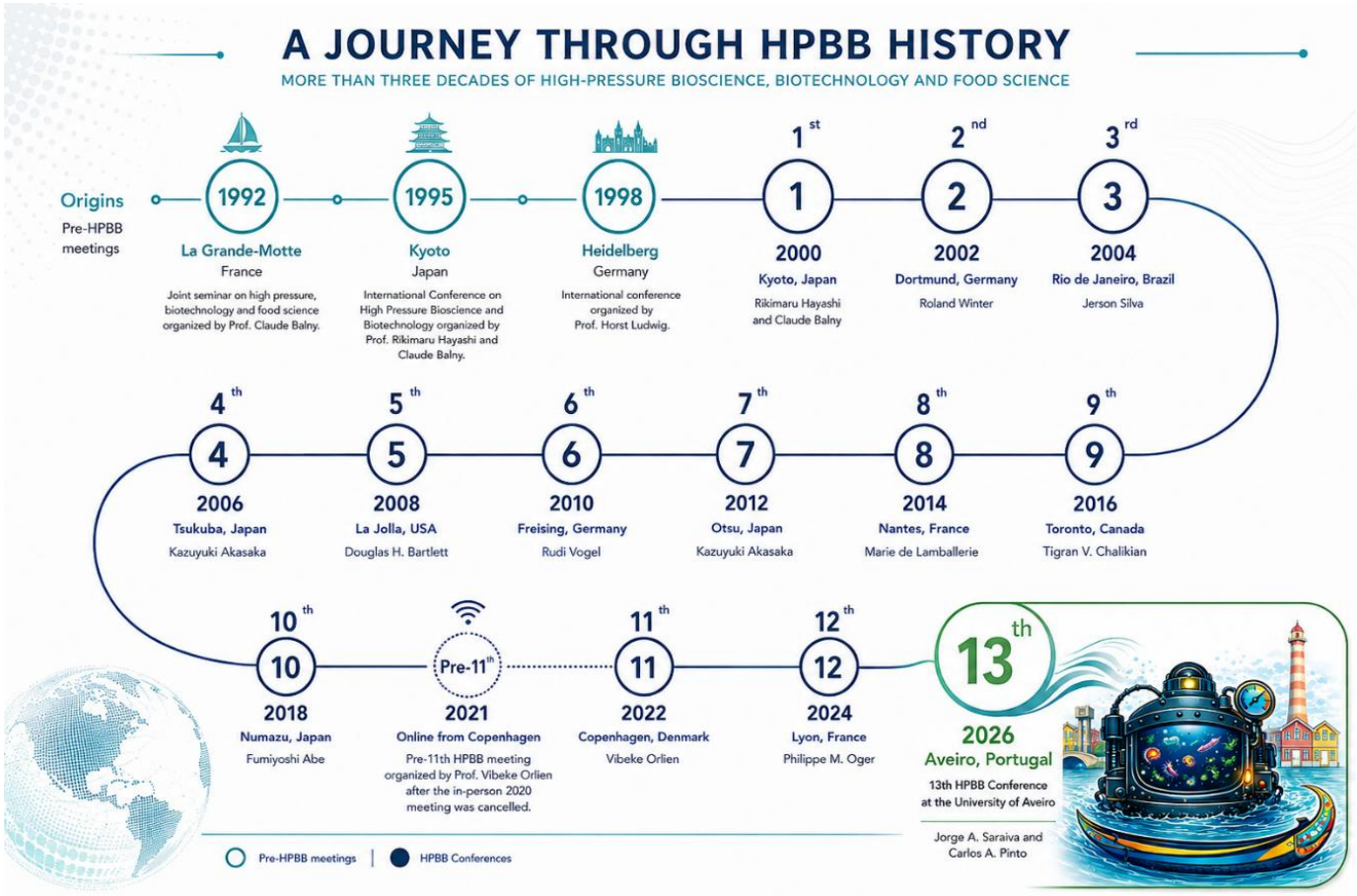
- **Vibeke Orlien**, University of Copenhagen, Denmark

HPBB - THREE DECADES OF SCIENTIFIC EXCHANGE

A brief history of the HPBB conference

HPBB grew from international high-pressure seminars into a recurring forum connecting food science, bioscience and biotechnology. The series has travelled across Europe, Asia and the Americas, building a close and collaborative global community:



Gold Sponsor

The organizing committee would like to extend its heartfelt thanks to **Hiperbaric SA.**, a world-leading company in High Pressure Processing (HPP) technology located in Burgos, Spain, for its invaluable support as a Gold Sponsor of the 13th International Conference on High Pressure Bioscience and Biotechnology (HPBB 2026), taking place in Aveiro, Portugal, from 31st August - 3rd September 2026.

We are truly grateful for Hiperbaric's generous support and continued commitment to the high-pressure scientific and technological community. Your contribution is extremely valuable to us and plays an important role in making this Conference possible.

We would also like to express our sincere appreciation for the HPP-treated products kindly provided by Hiperbaric for the Welcome Coffee Break on August 31st. This is especially meaningful as the opening day of the conference will include a visit to the University of Aveiro's high-pressure facilities, together with demonstrations using our high-pressure equipment.

Having these products available during the Welcome Coffee Break will give participants the opportunity to experience first-hand some real-world applications of HPP technology, creating a perfect connection between the scientific programme, the equipment demonstrations, and the practical impact of high-pressure processing.

Once again, a heartfelt thank you to the entire Hiperbaric team for your generosity, your trust, and your support of HPBB 2026. We are deeply grateful to have Hiperbaric as our Gold Sponsor and as part of this special event in Aveiro.

Want to know more about Hiperbaric? Please visit the company's website: https://www.hiperbaric.com/en/

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WELCOME TO THE PORTUGUESE VENICE

Discover Aveiro

A compact coastal city shaped by its lagoon, Aveiro brings together canals, *Art Nouveau* façades, maritime traditions and the distinctive colours of nearby Costa Nova.

Aveiro channels and Moliceiros • Salt pans • Art Nouveau • Costa Nova

Canals & *Moliceiros* — Traditional painted *Moliceiro* boats animate the urban channels and recall the lagoon's working heritage.

***Art Nouveau* —** Ornamental façades and ceramic details give the historic centre a character that is best explored on foot.

Salt & lagoon — The salt pans and Ria de Aveiro landscape connect the city to a living coastal ecosystem and a long tradition of salt production.

Costa Nova — Striped fishermen's houses and the Atlantic shore add a vivid coastal counterpoint only a short distance from the city.

EXPERIENCE AVEIRO TOGETHER

Social programme

Two shared moments bring the conference community beyond the lecture hall and into the landscape and hospitality of Aveiro.

Traditional *Moliceiro* boat tour

A relaxed tour through Aveiro's urban channels, with views of the city's painted boats, Art Nouveau architecture and lagoon heritage.

Conference dinner · University of Aveiro Restaurant

The conference dinner will be held at the University of Aveiro restaurant, offering an informal setting for conversation and celebration within the university campus.

Conference Programme

August 31st (Monday)

17h00 – 20h00 **Registration**

Pre-meeting-welcome-get-together and visit to the high pressure facilities at the University of Aveiro (*includes degustation of several high pressure pasteurized foods and in loco showcasing demonstrations of high pressure applications)*

Rectory Central Building, University of Aveiro

Google Maps location: https://maps.app.goo.gl/zdSxMRjJpY6D2Cbv6

September 1st (Tuesday)

8h30 **Registration**

9h00 – 09h30 **Conference opening**

Jorge Manuel Alexandre Saraiva, Conference Chair, University of Aveiro

Artur Manuel Soares da Silva, Rector of the University of Aveiro

Armando Jorge Domingues Silvestre, Director of the Chemistry Department of the University of Aveiro

Fumiyoshi Abe, Aoyama Gakuin University, Japan, The International Association of High Pressure Bioscience and Biotechnology

Food Science under High Pressure – Session 1 — Chair: Kazutaka Yamamoto

09h30 – 10h15 **Keynote 1:** Pressure effects on non-enzymatic reaction kinetics in food systems: from degradation mechanisms to stabilization strategies

Avi Shpigelman (Technion, Israel)

10h15 – 10h45 **Coffee-break and Poster session**

Food Science under High Pressure – Session 2 — Chair: Carlos A. Pinto

10h45 – 11h05 **O1:** Watermelon juice preservation by moderate pressure pasteurization at room temperature

Vasco Lima (University of Aveiro, Portugal)

11h05 – 11h25 **O2:** Effect of high pressure processing on microbial inactivation and stability of strawberry tree juice during refrigerated storage

Enrique Pino-Hernández (TagusValley, Portugal)

11h25 – 11h45 **O3:** Validation of *Listeria innocua* UBU as a surrogate for *Listeria monocytogenes* in meat products treated by high pressure processing (HPP)

Rui Queirós (Hiperbaric SA., Spain)

11h45 – 12h05 **O4:** High pressure processing for milk safety and subsequent yogurt fermentation

Nicole Moreira (University of Aveiro, Portugal)

12h15 – 14h00 **Lunch**

University of Aveiro Restaurant

Google Maps location: https://maps.app.goo.gl/MYP8ZPMNYWetqAQ29

Food Science under High Pressure – Session 3 — Chair: Jorge A. Saraiva

14h00 – 14h20 **Golden sponsor presentation** - Commercial and Future Applications of High-Pressure Processing (HPP): From Global Market Expansion to Emerging Technologies
Carole Tonello-Samson (Hiperbaric SA., Spain)

14h20 – 14h40 **O5:** Effect of NaCl on the syneresis of egg white gel solidified by high hydrostatic pressure
Kazutaka Yamamoto (Institute of Food Research, NARO, Japan)

14h40 – 15h00 **O6:** Impact of High Pressure Processing Pre-treatments on Physicochemical, Sensory and Microbiological Quality of Freeze-Dried Beetroot Snacks
Telma Orvalho (TagusValley, Portugal)

15h00 – 15h20 **O7:** Effects of two pressure-based pasteurizations and packaging atmosphere on lipid oxidation of smoked salmon
Alireza Mousakhani Ganjeh (University of Aveiro, Portugal)

15h30 – 16h00 **Coffee-break and Posters session**

Food Science under High Pressure – Session 4 — Chair: Carole Tonello-Samson

16h00 – 16h20 **O8:** Changes in Quality-Related Compounds in Purple Carrot Juice after High-Pressure Thermal Sterilization Treatments by PESI-MS/MS Analysis
Miho Kakui (Kakui Food Co.,LTD, Japan)

16h20 – 16h40 **O9:** Effect of HHP treatment for the preservation of the olive pomace of the Morisca cultivar as a food additive
María Cabeza de Vaca (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

16h40 – 17h00 **O10:** Effects of sodium chloride and osmotic conditions on the barotolerance of *Staphylococcus aureus* under high hydrostatic pressure treatment
Satomi Tsutsuura (Niigata University, Japan)

17h00 – 18h30 **Sunset refreshment cocktail**
Rectory Central Building, University of Aveiro
Google Maps location: https://maps.app.goo.gl/zdSxMRjJpY6D2Cbv6

September 2nd (Wednesday)

8h30 **Registration**

Cells, Organisms and Biotechnology Applications under High Pressure – Session 1
Chair: Roland Winter

9h00 – 09h45 **Keynote 2:** Activation of cell motility measured by high-pressure microscopy
Masayoshi Nishiyama (KINDAI University, Japan)

09h45 – 10h05 **O11:** Room temperature hyperbaric storage of human adipose derived stem cells as a potential alternative to cryopreservation
Ana P. Martins (University of Aveiro, Portugal)

10h15 – 10h45 **Coffee-break and Poster session**

Cells, Organisms and Biotechnology Applications under High Pressure – Session 2
Chair: Werner Kremer

10h45 – 11h05 **O12:** Development of high pressure transformation of the haloalkaliphilic archaeon *Natronomonas pharaonis*
Emma Bonnaud (INSAVALOR, France)

11h05 – 11h25 **O13:** Membrane adaptation to high pressure involves bulk lipid transfer at ER-PM contact sites
Fumiyoshi Abe (Aoyama Gakuin University, Japan)

11h25 – 11h45 **O14:** Expanding the Pressure Frontier in Gruneisen Parameter Measurement
Jun Kong (Center for High Pressure Science and Technology Advanced Research, China)

11h45 – 12h05 **O15:** High pressure treatment of cellular assemblies towards stable therapeutic products Mariana Oliveira *(University of Aveiro, Portugal)*

12h05 – 12h15 **Conference photo**
Rectory Central Building, University of Aveiro

12h30 – 14h00 **Lunch**
University of Aveiro Restaurant
Google Maps location: https://maps.app.goo.gl/MYP8ZPMNYWetqAQ29

15h00 – 16h15 **Social programme**
Traditional *Moliceiro* boat tour through the channels of Aveiro
Google Maps Location: https://maps.app.goo.gl/iwUUG2RLBDdzpqKv6

SCAN OR CLICK ME

16h15 – 19h00 **Free time**
Explore the beautiful city of Aveiro (touristic spots in the QRCODE)

20h00 – 23h00 **Conference dinner**
University of Aveiro Restaurant
Google Maps location: https://maps.app.goo.gl/MYP8ZPMNYWetqAQ29

September 3rd (Thursday)

Structure, Dynamics and Function of Biomacromolecules under High Pressure – Session 1

Chair: Masayoshi Nishiyama

09h00 – 09h45 **Keynote 3:** Biomolecules under Stress - Life under Extreme Conditions
Roland Winter (Technical University of Dortmund, Germany)

09h45 – 10h05 **O16:** Insights into the Structure of Invisible Conformations of Large Methyl Group Labelled Molecular Machines from High Pressure NMR
Werner Kremer (University of Regensburg, Germany)

10h15 – 10h45 **Coffee-break and poster session**

Structure, Dynamics and Function of Biomacromolecules under High Pressure – Session 2

Chair: Fumiyoshi Abe

10h45 – 11h05 **O17:** Influence of high hydrostatic pressure pretreatment on the extractability and composition of citrus pectin
Brian Macias Frotto (Tecnológico de Monterrey, Mexico)

11h05 – 11h25 **O18:** Adaptation to high salinity: Response of bacterial model membranes to Martian deep subsurface conditions
Attila Tortorella (TU Dortmund University, Germany)

11h30 – 11h45 **Presentation of next HPBB**

11h45 – 12h00 **Conference closing and farewell**

Keynote Lectures

Food Science under High Pressure

Cells and Organisms and Biotechnology Applications under High Pressure

Structure, Dynamics and Function of Biomacromolecules under High Pressure

Gold Sponsor Communication

Keynote lectures

K1 - Food *Science under High Pressure*

Pressure effects on non-enzymatic reaction kinetics in food systems: from degradation mechanisms to stabilization strategies

Avi Shpigelman, Technion, Israel

K2 - *Cells and Organisms and Biotechnology Applications under High Pressure*

Activation of cell motility measured by high-pressure microscopy

Masayoshi Nishiyama, KINDAI University, Japan

K3 - *Structure, Dynamics and Function of Biomacromolecules under High Pressure*

Biomolecules under Stress – Life under Extreme Conditions

Roland Winter, Technical University of Dortmund, Germany

Gold Sponsor Communication

G1 - Food Science *under High Pressure*

Commercial and Future Applications of High-Pressure Processing (HPP): From Global Market Expansion to Emerging Technologies

C. Tonello-Samson, Hiperbaric SA, Burgos, Spain

Hiperbaric
HIGH PRESSURE TECHNOLOGIES

Pressure effects on non-enzymatic reaction kinetics in food systems: from degradation mechanisms to stabilization strategies

Or Shapira[1], Rachel Levy[1], Hani Shkolnikov Lazober [1], Eden Eran Nagar[1], Inbal Hanuka[1], Valeria Weiss[1], Zoya Okun[1], Avi Shpigelman[1,2,3]

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[2] *Hecht Sustainable Protein Research Center, Technion, Israel Institute of Technology, Haifa 3200003, Israel*
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Hyperbaric storage (HS) is receiving increasing attention as a pressure-based alternative to conventional refrigeration, and its preservation performance is known to depend not only on pressure and storage time but also on matrix-related variables such as pH and water activity[1]. In parallel, understanding how pressure affects non-enzymatic reaction kinetics is essential for the rational design of high-pressure food preservation technologies, particularly when pressure is applied for prolonged periods (HS) or combined with thermal load. Such understanding is also relevant beyond HS, including other pressure-based processing and preservation technologies. In contrast to temperature, pressure may accelerate, decelerate, or negligibly affect a reaction, depending on the activation volume of the rate-determining step. However, direct pressure effects are often superimposed by co-occurring pressure-induced pH shifts, which remain insufficiently considered despite their mechanistic importance [2,3]. Furthermore, in complex food systems, pressure can also modify interactions among food components, thereby affecting structure, stability, and functionality.

In this talk, we will examine pressure effects on food compounds in model and multicomponent systems, with emphasis on polyphenols and phycobiliproteins. Using epigallocatechin gallate as an initial model, followed by catechins and anthocyanins under HS-like conditions, we show that in neutral baroresistant systems, pressure enhances degradation and yields negative activation volumes [2,3]. Extending beyond the single-compound model, structurally different catechins and anthocyanins exhibited markedly different intrinsic degradation rates, yet no significant structure-dependent effect of pressure on activation volume was observed within each subgroup, suggesting subgroup-dependent mechanistic responses to pressure (**Figure 1**). These observations were further supported in multicomponent extract systems, indicating that the pressure effect is retained beyond isolated purified compounds.

On the other hand, in non-baroresistant systems, pressure-induced pH shifts can partially offset or, at lower pressures, overcome the direct accelerating effect of pressure, highlighting the need to distinguish between intrinsic pressure effects and indirect medium/matrix effects [2,3]. In contrast, recent work on glucose-glycine model systems under HS showed pressure-dependent suppression of Maillard development, emphasizing that pressure does not exert a universal protective or detrimental effect on food reactions [4]. Pressure-induced pH shifts may also contribute to other pressure-mediated phenomena, including protein gelation.

To move closer to real foods, we will also discuss how matrix interactions affect stability and how pressure-modified food structures may influence downstream functionality, including gelation behaviour and, in selected cases, bioaccessibility.

To broaden this perspective beyond flavonoids, we also discuss related kinetic observations on phycocyanobilin and phycobiliproteins. In these systems, phycobiliprotein color deterioration rates under high pressure were lower than under thermal treatment, whereas isolated phycocyanobilin remained stable during processing but oxidized during storage in a pH-dependent manner [5].

Together, these results support a reaction-specific and matrix-aware view of pressure effects in foods and highlight the importance of considering both activation volume and pressure-induced pH shifts when developing robust high-pressure preservation strategies.

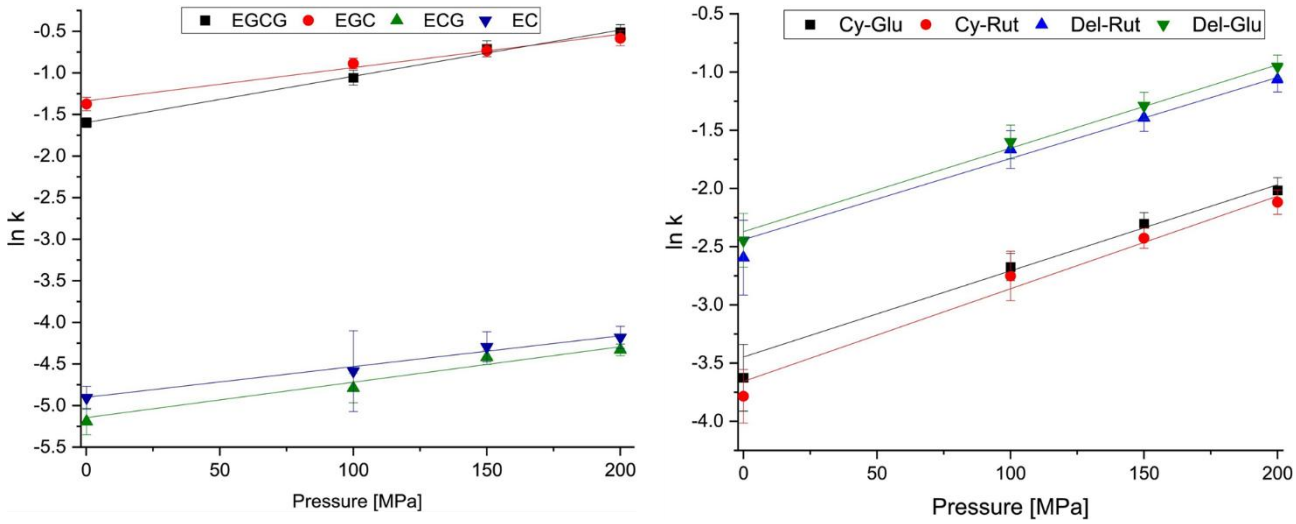

Figure 1. Dependence of ln k on pressure for selected Catechins (left) and anthocyanins (right) under hyperbaric storage-like conditions in a neutral baroresistant buffer. The approximately linear increase in ln k with pressure indicates negative activation volumes, while similar slopes within each subgroup suggest shared pressure dependence despite structural diversity.

Acknowledgements and funding

This research was funded by the Israel Science Foundation, grant number 685/17. Rachel Levy is grateful to the Azrieli Foundation for the award of an Azrieli Fellowship. Work related to phycocyanobilin and phycobiliproteins was partially funded by the ELITE FOOD ENGINEERING RESEARCH FUND 1017810. Part of the work was supported by Niedersachsen-Israel-Cooperation grant ZN3170. Part of the work was supported by the Laura Gurwin Flug Family Fund. Part of the work was supported by grant (no. 3-11656) from the Israeli Ministry of Health, Chief Scientist Office.

References

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Activation of cell motility measured by high-pressure microscopy

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High-pressure microscopy is one of the powerful techniques to visualize the effects of hydrostatic pressures on research targets [1]. It could be used for monitoring the pressure-induced changes in the structure and function of molecular machines in *vitro* and *in vivo*. We developed a high-pressure chamber for optical microscopy (**Figure 1a and b**). This chamber was optimized for both the best image formation and for the stability to hydrostatic pressure up to 150 MPa. This maximum pressure value using this microscope is ~1.5-fold higher than the hydrostatic pressure in the deepest part of the Mariana Trench (~10,900 m in depth). The similar chamber is commercially available (Syn-corporation, Kyoto, Japan). The high-pressure chamber and hand pump were combined with an inverted microscope (Ti-*E*, Nikon, Japan). By using the developed system, we studied the pressure response of various biological samples such as molecular machines, cells and individuals [2-4].

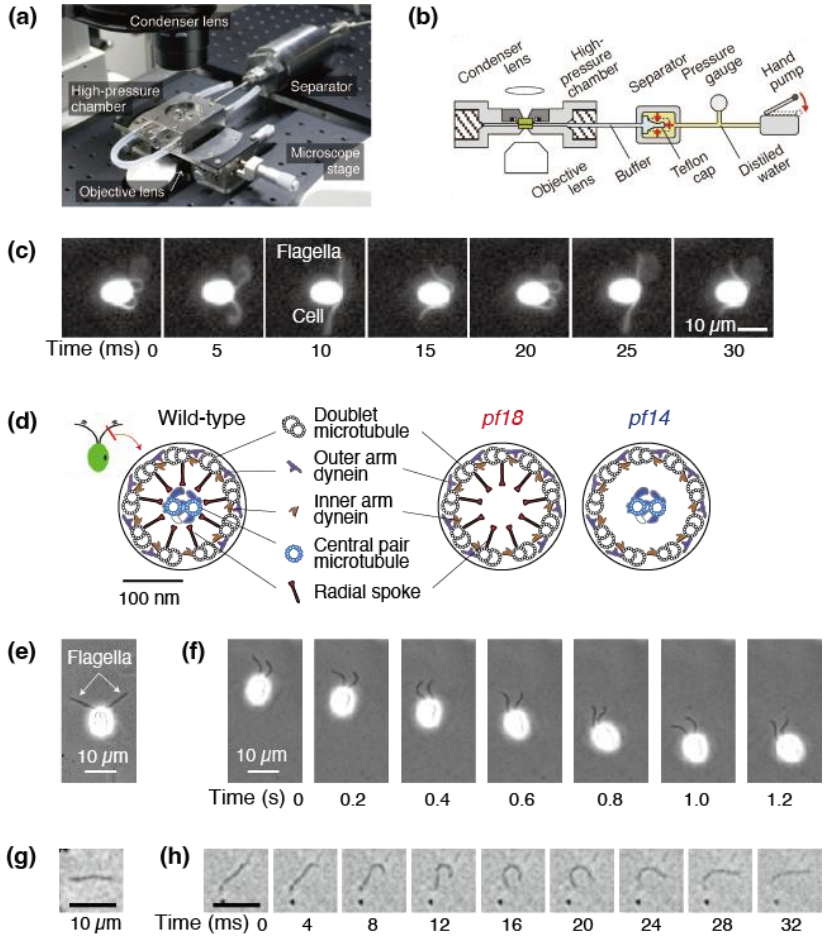

Figure 1. Pressure-induced activation of flagellar beating in *Chlamydomonas* cell. (a) High-pressure chamber for optical microscopy. (b) Schematic drawing of the experimental setup (not to scale) [1]. (c) Snapshots of a swimming wild-type cell. (d) Schematic drawing of the flagellum in cross section. (e and f) Phase-contrast image of *pf*18 cells at 0.1 (e) and 80 MPa (f). (g and h) Phase-contrast image of *pf*14 axonemes at 0.1 (g) and 40 MPa (h).

Here, we report two types of flagellar motility at high-pressure. Eukaryotic flagella and cilia generate rhythmical beating motions. Wild-type *Chlamydomonas* cells swim smoothly in solution by using two flagella to stroke in a breaststroke-like motion (**Figure 1c**). The beating of eukaryotic flagella depends on the sliding movements between microtubules powered by dynein. In cilia/flagella of most organisms, microtubule sliding is regulated by the internal structure of cilia comprising the central pair (CP) of microtubules and radial spokes (RS) (**Figure 1d).** We examined the motility of wild type and two kinds of non-motile paralyzed-flagella (*pf*) mutants, *pf*18 lacking CP and *pf*14 lacking RS (**Figure 1d**) [5]. The *pf*18 mutant was non-motile under physiological conditions (**Figure 1e**). On the other hand, when the pressure was increased to 80 MPa, many *pf*18 cells began swimming within a few seconds (**Figure 1f**). Similarly, the *pf*14 mutant also began swimming in solution under high-pressure conditions. Similar results were also obtained from *pf*14 mutant. The activation of flagellar motility in the *pf* mutants may be brought about through a direct action of pressure on the axoneme. Alternatively, motility may be induced through some vital cellular function. To distinguish between the two possibilities, we performed *in vitro* assays at high pressures using isolated axonemes. The isolated *pf*14 axoneme displayed no movements at 0.1 MPa (**Figure 1g**), but vigorous beating motions at 40 MPa (**Figure 1h**). Similar results were also obtained from *pf*18 mutant. The observation strongly suggest that the applied pressure induced flagellar movements in *pf* cells by directly acting on the axoneme. The isolated flagella beating pattern in *pf*18 and 14 mutants at high-pressure was similar to that of wild type at 0.1 MPa. In addition, at 80 MPa, flagella underwent an asymmetric-to-symmetric waveform conversion, similar to the one triggered by an increase in intra-flagella Ca^{2+} concentration during cell's response to strong light. These results showed that neither beating nor waveform conversion of cilia/flagella requires the presence of CP/RS in the axoneme.

Next, we measured the swimming motility of non-motile sperm cells in a knockout mouse model. The cells lacked a part of radial spoke 1 (RS1) in sperm flagella (**Figure 1d**), due to the deletion of *Iqub* [6]. At ambient pressure (0.1 MPa), the sperm cells were unable to swim in solution with weak flagellar motility. When the pressure was increased to 60 MPa, a certain number of the cells started to swim unidirectionally in solution. After the pressure release, all the cells stopped swimming. Thus, the pressure-induced activation of the flagellar motility was reversible. This result is consistent with previous results in *Chlamydomonas pf* mutants [5]. We will discuss the experimental results in detail.

Acknowledgements and funding

This work was financially supported by the Japan Society for the Promotion of Science (JSPS KAKENHI; Grant No. JP23K26497 and JP25K01633 to MN).

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K3 - Structure, Dynamics and Function of Biomacromolecules under High Pressure

Biomolecules under Stress – Life under Extreme Conditions

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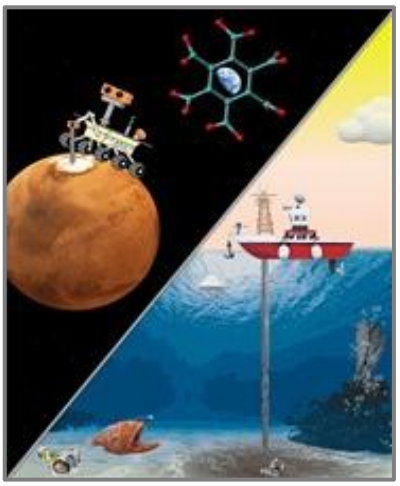
Elucidating the details of the formation, stability, interactions, and reactivity of biomolecular systems under extreme environmental conditions, including high and low temperature, high salt concentrations in brines, high osmotic as well as high hydrostatic pressure, is of fundamental biological, astrobiological and biotechnological importance. Bacteria and archaea are able to survive in the deep ocean or in the subsurface of the Earth, where pressures up to more than 1000 bar are reached. The deep subsurface of Mars may harbor high concentrations of salts in brines, such as aggressive perchlorates, but we know little about how these conditions and the resulting osmotic and hydrostatic stress conditions would affect the habitability of such environments for cellular life. By combining various biophysical techniques, including calorimetry, confocal fluorescence microscopy, fluorescence, smFRET, UV/Vis, CD, FTIR, and NMR spectroscopy as well as small-angle X-ray and neutron scattering, the combined effects of temperature, osmotic and hydrostatic pressure and cosolutes on the structure, solvational properties, dynamics and intermolecular interactions of proteins, nucleic acids, and lipid membranes of different complexity have been investigated [1-6]. Pressure-jump relaxation studies have been used to study the kinetics and mechanisms of biomolecular phase transitions and structural transformations, such as membrane fusion or protein and nucleic acid folding. High pressure kinetics experiments have been carried out to reveal the effects of pressure on ligand binding and reactions of enzymes. Selected results are presented [1-6].

A particular focus is on the combined effects of temperature, pressure and cosolvents on assembly processes based on liquid-liquid phase separation (LLPS) phenomena of biomolecular systems, as well as their effects on the temperature and pressure dependence of ligand binding and enzymatic reactions. Knowledge of these effects is essential for our understanding of life exposed to harsh environmental conditions and of the physical limits of life in general. We have studied various systems that undergo LLPS, including lysozyme, crystallin, elastin, LAF1, FUS, insulin-derived peptides, and proteins that mimic postsynaptic densities [7-18]. Strikingly, many biomolecular condensates are more pressure sensitive than protein unfolding, suggesting that organisms living in the deep sea and in sediments beneath the seafloor must mitigate this pressure sensitivity to avoid undesirable destabilization of their functional biomolecular condensates. We have found that crowding and certain deep sea osmolytes can stabilize protein droplets under pressure.



Apart from playing the well-recognized roles in the self-assembly of intracellular machinery, LLPS constitutes a hypothetical, yet compelling scenario for prebiotic compartmentalization. One challenge facing this idea is the scarcity of typical biopolymer precursors of liquid droplets on early Earth. We demonstrate that mellitic acid, an abiotic component found in certain minerals and a marker of organic matter in astrobiology, forms liquid droplets with poly-L-lysine across a broad pH, temperature and pressure range and various mixing ratios. We propose that the capacity to induce LLPS with short oligocations, along with abiotic synthesis routes, positions mellitic acid as a viable candidate for an ingredient of early prebiotic membraneless protocells [19].

Acknowledgements and funding

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HIGH PRESSURE TECHNOLOGIES

G1- Commercial and Future Applications of High-Pressure Processing (HPP): From Global Market Expansion to Emerging Technologies

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High-Pressure Processing (HPP) has been developed from a niche food preservation technology into a globally established industrial solution, with around 800 systems installed worldwide and continued double-digit market growth. Today, HPP is widely recognized for its ability to enhance food safety, extend shelf life, and preserve fresh-like quality while consumer demand for clean-label products is increasingly being met. The current commercial landscape of HPP is shaped by regional market trends, key application sectors, and emerging growth opportunities across North America, Europe, Latin America, Asia-Pacific, and the Middle East and Africa.

Commercial success has been demonstrated across diverse categories, including premium beverages, raw and fresh pet food, dairy and high-protein products, seafood, ready-to-eat meals, cured meats, tropical juices, and avocado-based products. International trade has also been facilitated by HPP through compliance with stringent food safety requirements, particularly regarding *Listeria monocytogenes* control.

Beyond established commercial uses, emerging research directions are being developed through pressure-based technologies. High-Pressure Thermal Processing (HPTP) is being evaluated as an alternative strategy for the sterilization of low-acid foods, along with pressure-assisted extraction of bioactive and flavor compounds, synergistic combinations of HPP with bacteriophages for targeted microbial control, valorization of dairy side streams through pressure-assisted protein fractionation, and in-bulk processing concepts designed to improve operational efficiency in beverage manufacturing.

Industrial case studies have been integrated with recent scientific advances, showing how HPP is being transitioned from a preservation technology to a broader processing platform capable of addressing challenges related to food safety, sustainability, ingredient functionality, waste valorization, and product innovation. HPP and related pressure-based technologies are expected to contribute to next-generation food processing systems.

Oral & Poster Communications

Food Science under High Pressure

Oral Communications

O1: *Watermelon juice preservation by moderate pressure pasteurization at room temperature*

Vasco Lima (Faculty of Biotechnology, Catholic University of Porto, Portugal)

O2: *Effect of high pressure processing on microbial inactivation and stability of strawberry tree juice during refrigerated storage*

Enrique Pino-Hernández (TagusValley, Portugal)

O3: *Validation of Listeria innocua UBU as a surrogate for Listeria monocytogenes in meat products treated by high pressure processing (HPP)*

Rui Queirós (Hiperbaric S.A., Spain)

O4: *High pressure processing for milk safety and subsequent yogurt fermentation*

Nicole Moreira (University of Aveiro, Portugal)

O5: *Effect of NaCl on the syneresis of egg white gel solidified by high hydrostatic pressure*

Kazutaka Yamamoto (Institute of Food Research, NARO, Japan)

O6: *Impact of high pressure processing pre-treatments on physicochemical, sensory and microbiological quality of freeze-dried beetroot snacks*

Telma Orvalho (TagusValley, Portugal)

O7: *Effects of two pressure-based pasteurizations and packaging atmosphere on lipid oxidation of smoked salmon*

Alireza Mousakhani Ganjeh (University of Aveiro, Portugal)

O8: *Changes in Quality-Related Compounds in Purple Carrot Juice after High-Pressure Thermal Sterilization Treatments by PESI-MS/MS Analysis*

Miho Kakui (Kakui Food Co., LTD, Japan)

O9: *Effect of HHP treatment for the preservation of the olive pomace of the Morisca cultivar as a food additive*

María Cabeza de Vaca (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

O10: *Effects of sodium chloride and osmotic conditions on the barotolerance of Staphylococcus aureus under high hydrostatic pressure treatment*

Satomi Tsutsuura (Niigata University, Japan)

Poster Communications

P1: *Application of High Pressure for Yogurt Preservation*

Beatriz Mota, Nicole Moreira, Carlos A. Pinto, Jorge A. Saraiva

P2: *High Hydrostatic Pressure Treatment on White Winemaking Lees (cv. Alarije)*

Ana María López Valenzuela (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P3: *Effect of High Hydrostatic Pressures on Red Winemaking Lees (cv. Tempranillo)*

Ana María López-Valenzuela (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P4: *High Hydrostatic Pressure (HHP) as a Preservation Strategy for Frankfurters Enriched with Grape Pomace*

M. J. Martín-Mateos (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P5: *Application of high-pressure hydrostatic technologies and skin packaging for the preservation of ready-to-eat lamb meat dishes*

M. J. Martín-Mateos (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P6: *Effect of HHP treatment for the preservation of the olive pomace of the Picual cultivar as a food additive*

María Cabeza de Vaca Molina (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P7: *Effect of HHP processing strategy (cycles vs continuous treatment) on colour and microbial stability of lamb burgers*

Miriam Sánchez Ordóñez (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P8: *Effect of high hydrostatic pressure on microbial inactivation and functional compounds in red pepper by-products*

Miriam Sánchez Ordóñez (Centro de Investigaciones Científicas y Tecnológicas de Extremadura, Spain)

P9: *Valorisation of chilli peppers by-products using high-pressure as an extraction technology*

Jéssica Tavares (University of Aveiro, Portugal)

P10: *Hyperbaric storage at room temperature of raw poultry meat as a novel preservation methodology compared to refrigeration*

Jéssica Tavares (University of Aveiro, Portugal)

P11: *Optimization of the extraction process of chilli peppers by-products using high-pressure*

Gabriela Matos (University of Aveiro, Portugal)

P12: *Preservation of egg white at room temperature by hyperbaric storage: impact on volatile profile and sensory attributes*

Gabriela Matos (University of Aveiro, Portugal)

P13: *Food Safety Assessment of Almond Ice Cream Formulated with Liquid Fresh Whey: Comparison between High Pressure Processing and Thermal Pasteurization*

Viviana Monteiro (TagusValley, Portugal)

P14: Antioxidant and antimicrobial activities of seaweed extracts obtained by high-pressure assisted extraction

Andreia Miranda (Instituto Politécnico de Leiria, Portugal)

Oral & Poster Communications

Cells and Organisms and Biotechnology Applications under High Pressure

Oral Communications

O11: *Room temperature hyperbaric storage of human adipose derived stem cells as a potential alternative to cryopreservation*

Ana P. Martins (University of Aveiro, Portugal)

O12: *Development of high pressure transformation of the haloalkaliphilic archaeon Natronomonas pharaonis*

Emma Bonnaud (INSAVALOR, France)

O13: *Membrane adaptation to high pressure involves bulk lipid transfer at ER-PM contact sites*

Fumiyoshi Abe (Aoyama Gakuin University, Japan)

O14: *Expanding the Pressure Frontier in Gruneisen Parameter Measurement*

Jun Kong (Center for High Pressure Science and Technology Advanced Research, China)

O15: *High pressure treatment of cellular assemblies towards stable therapeutic products*

Mariana Oliveira (University of Aveiro, Portugal)

Poster Communications

P15: *High hydrostatic pressure enhances cardiomyocyte contraction by regulating intracellular Ca^{2+} dynamics*

Masayoshi Nishiyama (KINDAI University, Japan)

Oral & Poster Communications

Structure, Dynamics and Function of Biomacromolecules under High Pressure

Oral Communications

O16: Insights into the Structure of Invisible Conformations of Large Methyl Group Labelled Molecular Machines from High Pressure NMR

Werner Kremer (University of Regensburg, Germany)

O17: Influence of high hydrostatic pressure pretreatment on the extractability and composition of citrus pectin

Brian Macias Frotto (Tecnológico de Monterrey, Mexico)

O18: Adaptation to high salinity: Response of bacterial model membranes to Martian deep subsurface conditions

Attila Tortorella (TU Dortmund University, Germany)

Oral communications and Posters

Food Science under High Pressure

O1: Watermelon Juice Preservation by Moderate Pressure Pasteurization at Room Temperature

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Watermelon juice (WJ) has an attractive colour, flavour, aroma and an interesting nutritional composition. However, it is very perishable because of its pH (5.2-6.7) and water activity (0.97-0.99) values, resulting in rapid microbial growth and enzymatic degradation[1], requiring effective preservation to ensure its safety and quality and extend its shelf-life.

Traditional thermal preservation methodologies are effective in reducing microbial loads, but they can also cause the loss of nutrients, bioactive compounds and affect the quality of the foods. So, nonthermal technologies have been increasingly studied and applied by the food industry. Pressure-based methodologies like high pressure processing are amongst the nonthermal technologies shown to be effective in assuring the safety of the food while having lower quality impact. Moderate pressure pasteurisation (MPP), which consists of storing foods under pressures usually up to 250 MPa for a few hours, has been studied for a potential pasteurization application at room temperature (RT) and studies have reported considerable microbial inactivation during MPP[2].

This work aimed at evaluating the impact of MPP (130-300 MPa) at RT (18-23 °C) for up to 24 hours on microbial (*Escherichia coli*, *Listeria monocytogenes* and *Saccharomyces cerevisiae*) and physicochemical parameters (colour, pH, total soluble solids, cloudiness and browning degree) of WJ (pH 6.5). The microbial inactivation kinetics were evaluated using the first-order linear model and the Weibull nonlinear model.

Results showed that MPP caused significant inactivation, often surpassing FDA's pasteurisation level of 5 log CFU/mL. MPP's effect on microbial counts depended on the microorganism and, especially, on the pressure level, since increasing the pressure resulted in higher inactivation rates. The microbial inactivation kinetic data was better fitted by the Weibull model, which allowed for more fits. MPP's impact on WJ's physicochemical parameters was overall limited to a decrease in cloudiness and minor colour variation.

These findings show that MPP can cause considerable inactivation (over 5 log CFU/mL) with limited physicochemical changes and without using high temperatures, thus having lower impact on organoleptic properties and energy costs, making MPP a possible environmentally friendlier process with great potential as a novel nonthermal pasteurization technique at RT.

Acknowledgements and funding

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O2: Effect of High Pressure Processing on Microbial Inactivation and Stability of Strawberry Tree Juice during Refrigerated Storage

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Escherichia coli is a Gram-negative microorganism widely associated with foodborne contamination, representing a significant safety concern in fruit-based products due to its ability to survive under acidic conditions and potentially recover during storage[1]. Despite its nutritional potential, strawberry tree (*Arbutus unedo* L.), a Mediterranean specie, remain underexploited due to its high perishability and handling limitations[2]. High pressure processing (HPP) has been applied as a nonthermal pasteurization technology, enabling effective microbial inactivation while preserving product quality. However, its efficiency depends on process conditions and matrix characteristics, requiring further investigation to ensure microbial stability during refrigerated storage[3].

This study aimed to evaluate the effectiveness of HPP on microbial inactivation and shelf-life stability of strawberry tree (*Arbutus unedo* L.) juice (pH 3.02 ± 0.01 and °Brix 14.50 ± 0.00) during refrigerated storage (5 ºC) for 30 days and compare it with thermal pasteurization. Experimental analyses were conducted on both inoculated samples with *Escherichia coli* (used as a target pathogenic surrogated microorganism to optimize the thermal and nonthermal pasteurization conditions) and on the juices' endogenous microorganisms (namely total aerobic mesophiles, psychrophiles and yeasts and moulds). HPP treatments (300-600 MPa for 5 min at room temperature), were applied and compared with conventional thermal pasteurization (70 ºC for 5-30 min). Additionally, challenge tests using pre-deteriorated (purposedly spoiled juice kept at room temperature for 16 days to induce microbial growth) were performed simulate a scenario of a juice heavily contaminated to assess the maximum microbial inactivation capacity of each treatment.

HPP treatments achieved more than 6-log units' inactivation of *E. coli* below detection limits under all tested conditions (300, 400, 500 and 600 MPa for 5 min, 18 ºC), similarly to thermal pasteurization (at 70 ºC for 5, 10, 20 and 30 min).

When it comes to the challenge tests with spoiled juice, the inactivation levels of total aerobic mesophiles and yeasts and moulds were directly proportional to the pressure level applied, *i.e.*, higher pressures (≥400 MPa for 5 min) resulted in higher microbial inactivation (with more than 6 log units' reduction), but still with a low residual fraction (**Figure 1A**), which remained within acceptable hygiene thresholds, while thermal pasteurization (70 ºC for 30 min) was unable to reach the 5-log units reduction (**Figure 1B**). This contrasts the feasibility of HPP to achieve higher microbial loads reductions in shorter processing times when compared to thermal pasteurization.

In natural juice (non-inoculated juice, containing only its indigenous fraction of microorganisms), all treatments (thermal pasteurization and HPP) initially reduced microbial loads to below detection limits. During 30 days of refrigerated storage, both thermal pasteurization and HPP (≥400 MPa) maintained microbial stability, with total aerobic mesophiles, psychrophiles and yeasts and moulds remaining below detection limits.

Overall, HPP proved to be an effective nonthermal alternative to conventional thermal pasteurization, ensuring both microbial safety and extended shelf-life in strawberry tree juice, in shorter processing times. These findings support the potential application of HPP in the preservation of high-value fruit juices.

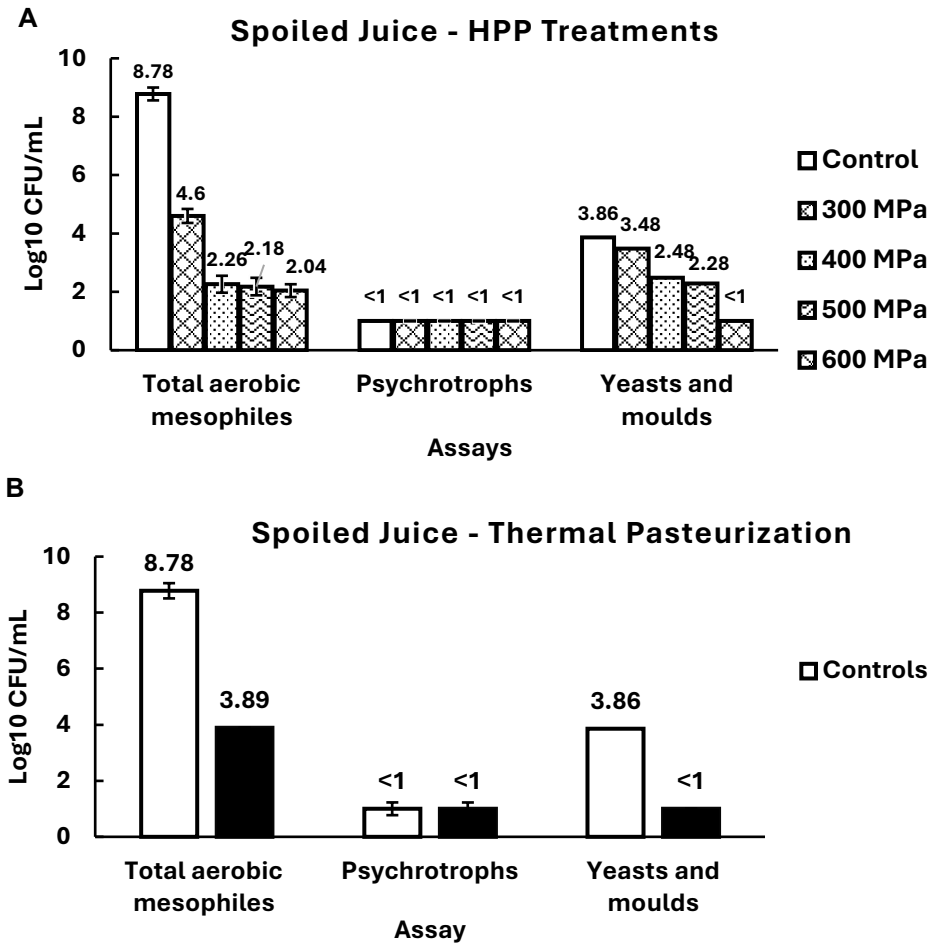

Figure 1. Microbial loads of total aerobic mesophiles, psychrophiles, and yeasts and moulds expressed as the decimal logarithm of colony forming units per mL of strawberry tree juice (Log_{10} CFU/mL) in spoiled juice (stored for 16 days at room temperature prior to processing), used to assess the impact of (A) high-pressure processing (300–600 MPa, 5 min, 18 ºC) and (B) thermal pasteurization (70 ºC, 30 min) on the inactivation of indigenous microorganisms.

Acknowledgements and funding: The authors gratefully acknowledge Medronho & Canela (Portugal) for providing strawberry tree fruits. This work received support from PT national funds (FCT/MCTES, *Fundação para a Ciência e Tecnologia and Ministério da Ciência, Tecnologia e Ensino Superior*) through the project UID/50006/2025, DOI 10.54499/UID/50006/2025 - Laboratório Associado para a Química Verde - Tecnologias e Processos Limpos (LAQV-REQUIMTE) and UNEDO4All - Estratégias inovadoras para conservação e valorização integral do Medronho na indústria alimentar (COMPETE2030-FEDER-02044600).

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O3: Validation of *Listeria innocua* UBU as a surrogate for *Listeria monocytogenes* in meat products treated by high pressure processing (HPP)

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Listeria monocytogenes remains one of the most important foodborne pathogens in ready-to-eat (RTE) foods due to its high severity and its persistence in food processing environments. High pressure processing (HPP) is increasingly used as a nonthermal preservation technology to improve the microbiological safety of meat products while maintaining their sensory quality[1,2]. However, industrial validation studies using pathogenic microorganisms are complex and involve biosafety concerns. For this reason, the use of a non-pathogenic surrogate with a response similar to that of *L. monocytogenes* is of practical interest for safe process validation[3].

This study evaluated the suitability of *Listeria innocua* UBU as a surrogate for *L. monocytogenes* in meat matrices subjected to HPP. This strain of *L. innocua* was compared with a cocktail of five *L. monocytogenes* strains selected according to pressure resistance, virulence and isolation source. Nine commercial meat products were studied: raw minced beef, cooked chicken, cooked ham, foie gras, cured loin, cured ham, chorizo, salchichón and salami. Samples were inoculated to initial levels of approximately 10^7 CFU/g, vacuum packaged and treated at 600 MPa for 3, 6 or 9 min at 25 °C. Microbial reductions were determined by plate count, and the influence of pH and water activity (a_w) on HPP inactivation was assessed.

Across all products, *L. innocua* showed an inactivation pattern like that of *L. monocytogenes*. Cooked and raw matrices showed the highest HPP lethality (from ~4 to >6 log CFU/g), fermented products showed intermediate reductions (from ~1 to 4.5 log CFU/g), and cured products were the most protective with the lowest reductions (from ~0.5 to 3 log CFU/g). Greater inactivation was associated with higher a_w and lower pH, confirming the important influence of product physicochemical properties on HPP lethality[4,5].

There were no significant differences in many cases between the surrogate and the pathogen. Where differences were detected, *L. monocytogenes* exhibited a higher level of inactivation compared to *L. innocua*, indicating greater inactivation of *L. monocytogenes* and thus a more conservative response of *L. innocua*. The largest effects were observed in chorizo (up to 1.97 log CFU/g at 9 min; $p < 0.001$), with smaller but significant differences in cooked chicken (0.39 – 0.83 log CFU/g; $p < 0.01$) and cooked ham (0.19 – 0.41 log CFU/g; $p < 0.05$) – **Table 1**. In fermented products, *L. innocua* was generally more resistant than *L. monocytogenes*, which is advantageous from a validation perspective because the inactivation of the surrogate would ensure at least the same, or greater, inactivation of the pathogen.

Table 1. Average difference in $\log_{10}$ inactivation between *L. monocytogenes* and *L. innocua*, calculated as *L. monocytogenes* minus *L. innocua*, across three HPP treatment durations (3, 6 and 9 min) at 600 MPa. Positive values indicate that *L. monocytogenes* was inactivated more than *L. innocua*. Statistically significant differences ($p < 0.05$) are highlighted in green.

	Holding time at 600 MPa			
Product	**3 min**	**6 min**	**9 min**	
Chorizo	1.07 ± 0.27	1.28 ± 0.58	1.97 ± 0.49	p < 0.001
Cooked chicken	0.63 ± 0.47	0.39 ± 0.20	0.83 ± 0.45	p < 0.01
Cooked ham	0.41 ± 0.26	0.36 ± 0.20	0.19 ± 0.43	p < 0.05
Cured loin	-0.26 ± 0.30	-0.03 ± 0.92	0.24 ± 0.94	p ≥ 0.05
Dry-cured ham	0.66 ± 0.56	1.03 ± 0.63	0.50 ± 1.29	
Foie gras	0.06 ± 0.55	-0.01 ± 0.82	0.08 ± 0.66	
Minced beef	0.23 ± 0.73	0.64 ± 0.58	-0.29 ± 0.14	
Salami	0.56 ± 0.51	1.43 ± 0.47	1.46 ± 0.28	
Salchichón	0.75 ± 0.16	2.00 ± 0.33	1.78 ± 0.77	

Linear regression analysis of the complete dataset showed a significant relationship between the inactivation of *L. innocua* and *L. monocytogenes* (R^2 = 0.8255), indicating a strong overall correlation between both responses under the tested HPP conditions. Overall, the results support the use of *L. innocua* UBU as a suitable surrogate for *L. monocytogenes* in HPP validation studies of RTE meat products and confirm the potential of HPP as an effective intervention to improve the safety of a wide range of meat matrices.

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O4: High Pressure for Milk Safety and Subsequent Yogurt Fermentation

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High pressure processing (HPP) is a well-established nonthermal technology operating at 400–600 MPa based on isostatic and *Le Chatelier's* principles [1,2]. In yogurt manufacturing, conventional thermal pasteurization (TP) of milk requires intense conditions (typically 92 °C for 2 min) to ensure microbial safety, which can degrade heat-sensitive nutrients, resulting in quality loss [3,4]. Exploring HPP as an alternative is highly promising, as high pressure can achieve pasteurization while simultaneously promoting protein denaturation and structural modifications at low temperatures [1,5]. This study evaluated HPP as a nonthermal alternative to traditional TP for milk in yogurt manufacturing, focusing on microbial safety and subsequent fermentation kinetics.

First, a pathogen challenge test was performed on raw milk inoculated with *Salmonella enterica* and *Staphylococcus aureus* to compare two HPP treatments: 550 MPa/10 min versus 600 MPa/8 min. While both treatments inactivated *S. enterica* below the detection limit (< 2.7 Log CFU/g), *S. aureus* exhibited higher barotolerance. However, the 600 MPa/8 min treatment achieved a substantially higher reduction of *S. aureus* (3.18 Log CFU/g) compared to 550 MPa/10 min (1.59 Log CFU/g), leading to its selection for subsequent yogurt production. Regarding endogenous microbiota in non-inoculated milks, high initial counts were observed in raw milk for psychrotrophic bacteria (5.67 ± 0.00 Log CFU/g), yeasts and molds (4.13 ± 0.05 Log CFU/g) Enterobacteriaceae (4.43 ± 0.01 Log CFU/g), *S. enterica* (4.11 ± 0.00 Log CFU/g), *S. aureus* (4.43 ± 0.08 Log CFU/g), *Streptococcus thermophilus* (5.66 ± 0.07 Log CFU/g) and *Lactobacillus delbrueckii* subsp. *bulgaricus* (5.30 ± 0.00 Log CFU/g). Both TP and HPP milks successfully showed pathogens, yeasts/molds, and native LAB below the detection limit. Notably, HPP milk demonstrated superior performance over TP milk in controlling psychrotrophic bacteria: while TP showed psychrotrophic population at quantification limit (3.70 ± 0.00 Log CFU/g), HPP achieved inactivation below detection limits (< 2.7 Log CFU/g), suggesting a more effective hurdle against heat-resistant or spore-forming psychrotrophs.

TP and HPP milks were subsequently inoculated with a commercial starter culture and fermented at 37, 40, and 43 °C until reaching pH 4.6. As observed in **Figure 1**, higher temperatures accelerated the fermentation process, with HPP milk consistently exhibiting faster fermentation kinetics than TP milk across all conditions. Specifically, fermentation times for HPP versus TP milk were ~6h versus ~9h (at 37 °C), ~5h versus ~8h (at 40 °C), and ~ 5h versus ~7h (at 43 °C).

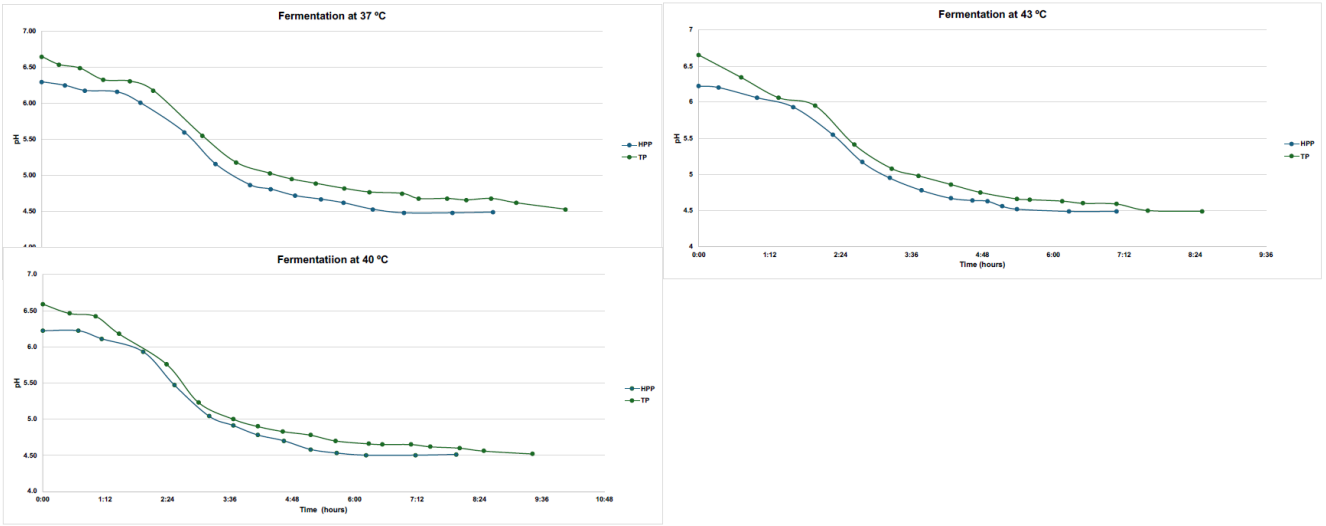

Figure 1. pH evolution throughout yogurt fermentation with HPP milk and TP milk at 37 ºC, 40 ºC and 43 ºC.

This acceleration in HPP milk was probably promoted by its lower initial pH (6.22–6.30) compared to TP milk (6.59–6.65) and different lag phase duration. At the end of fermentation, viable starter cultures (*Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *bulgaricus*) exceeded 9.00 Log CFU/g under all conditions (**Figure 2**), conferring the status of yogurt (> 7.00 Log CFU/g).

In conclusion, HPP at 600 MPa/8 min ensures microbial safety against key foodborne pathogens and endogenous spoilage microbiota, while considerably improving fermentation efficiency. These findings highlight HPP as a high-performing, sustainable technology for innovation in the dairy industry.

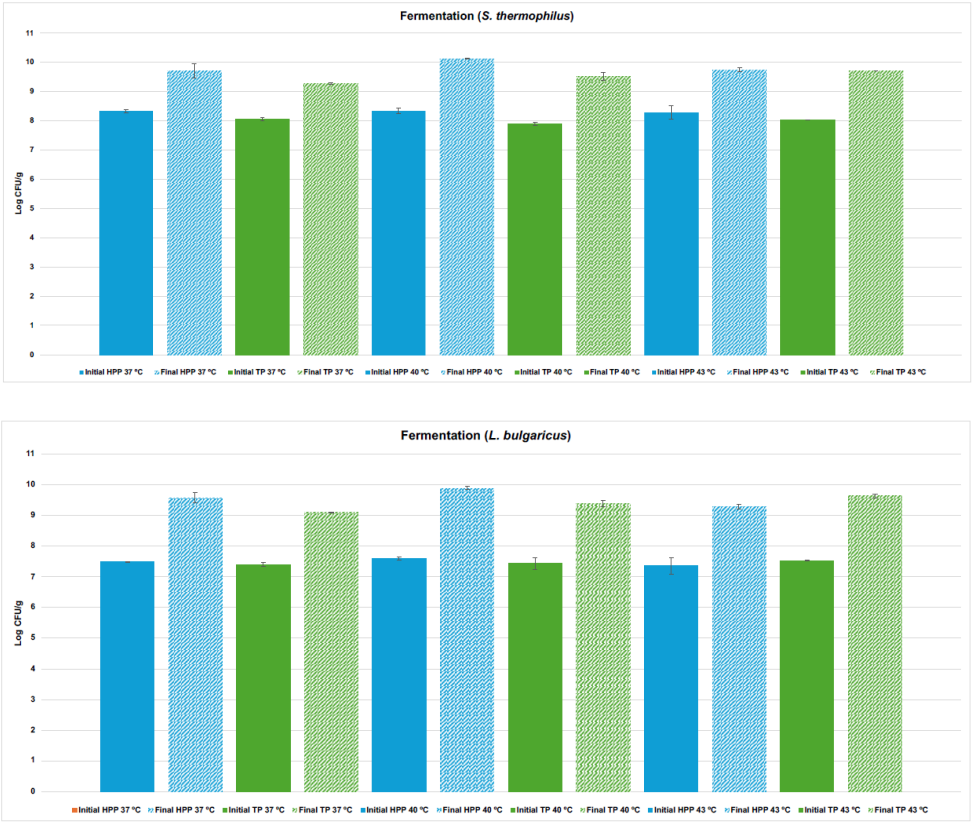

Figure 2. Initial (post-inoculation) and final (yogurt, pH 4.6) counts of *Streptococcus thermophilus* and *Lactobacillus delbrueckii* subsp. *Bulgaricus* (CFU/g) for fermentations with HPP milk and TP milk at 37, 40 and 43 ºC.

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O5: Effect of NaCl on the syneresis of egg white gel solidified by high hydrostatic pressure

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High hydrostatic pressure (HHP) has been commercially applied in food processing since 1990, when the first HHP-processed products were introduced in Japan. Since then, HHP at 500 - 600 MPa has been primarily used for cold pasteurization of juices and meat products. In addition, HHP can also induce starch gelatinization, protein denaturation, shucking, and liquid impregnation. However, industrial applications that exploit these effects, other than pasteurization, have remained limited to date [1].

HHP treatment induces egg solidification due to protein denaturation. Egg white denaturation was first reported by Dr. Percy William Bridgman, a Nobel Laureate in physics and a pioneer of high pressure research [2]. Since then, intensive research on individual egg proteins in egg white have been intensively carried out in the field of high pressure bioscience. In 1989, Dr. Hayashi, considered the father of HHP food processing, described solidification of whole egg in a Japanese book ”食品への高圧利用 (Application of High Pressure to Food)” [3]. However, egg white as a food material has not been sufficiently studied for industrial applications of HHP-solidified gel. In this study, egg white was subjected to HHP treatment at 600 MPa, and the properties of the resulting HHP-solidified egg white were investigated to evaluate its potential for commercial application.

Egg white was sealed in a dialysis tube, which was then placed in a plastic pouch containing 0–10 % NaCl solution prior to processing. HHP treatment was carried out using HHP processors (Dr. Chef, Kobe Steel Ltd., Kobe, Japan; TFS56-50, Toyo Koatsu, Co. Ltd., Hiroshima, Japan). As a conventional cooking method, heat treatment was conducted by boiling (100 °C, 10 min). Both HHP and heat-treated samples were stored at 5 °C and then subjected to quantitative evaluation.

For quantitative evaluation, changes in the weight of egg white samples were measured at 0, 24, 48, and 72 h. In addition, sample shape was evaluated using a laser-based three-dimensional scanner (SOL, Scan Dimension, Denmark). Shape parameters such as height, area (projected to the platform plate), and volume of the samples were then computed from the point-cloud data using custom software developed in Python 3, which read the data, separated the plate from the sample, and calculated these parameters. Texture profile analysis was conducted using a TA-XT Plus analyser (Stable Micro Systems, UK), in which cylindrical samples (30 mmϕ × 12 mm) were subjected to a double-bite test. Several plungers were used to characterize differences in the sample texture. Liquid holding capacity (LHC) was determined by centrifuging approximately 10 g of the gel samples at 10,000 × *g* at 5 °C for 20 min.

HHP-treated egg white released some liquid and with a visibly reduced volume; this phenomenon was identified as syneresis. During storage, HHP-treated samples exhibited liquid release and noticeable reductions in volume and weight, reflecting syneresis that intensified with increasing NaCl concentration. In contrast, heat-treated samples remained stable, with no visible syneresis. A reduction in LHC was observed with increasing NaCl concentration, and the reduction was pronounced for HHP-treated samples. Quantitative 3D data analysis showed that heat-treated samples underwent much smaller changes in weight and shape parameters, indicating little syneresis of boiled samples. By comparison, HHP-treated samples showed marked decreases of them after 1 day of refrigerated storage. The double-bite test indicated that heat-treated samples exhibited comparable first and second peak heights, whereas HHP-treated samples showed a higher second peak than the first, consistent with syneresis that depended on NaCl concentration. These results suggest that the double-bite test may be used to evaluate egg white syneresis based on differences between the peak heights.

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O6: Impact of Pulsed Electric Fields and High Pressure Processing Pre-treatments on Physicochemical, Sensory, and Microbiological Quality of Freeze-Dried Beetroot Snacks

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Freeze-drying is widely recognized as one of the most effective dehydration methods for preserving the organoleptic properties and nutritional value of fruits and vegetables when compared to conventional drying techniques. However, the high energy consumption resulting from long processing times constrains its application at an industrial scale[1]. In this context, the development of assisted freeze-drying processes using emerging technologies has gained increasing attention. Nonthermal approaches such as Pulsed Electric Fields (PEF) and High-Pressure Processing (HPP) have demonstrated potential as a pretreatment to enhance drying efficiency, promote tissue permeabilization and induce structural modifications that facilitate mass transfer, while simultaneously preserving or even improving the quality and sensory attributes of the final products[2,3]. These structural modifications are essential for enhancing flavour incorporation, enabling the efficient infusion of solutes into the vegetable matrix. Consequently, these strategies are especially relevant in light of the growing demand for high-quality vegetable snacks within more sustainable food systems, contributing to the development of value-added dehydrated products[1,2].

The present study aimed to evaluate the effect of nonthermal emerging processing technologies, namely PEF and HPP, as pre-treatments in the development of a freeze-dried beetroot snack produced from low-caliber beetroot. The impact of these approaches on the physicochemical, sensory, and microbiological properties of the final product was assessed, while simultaneously targeting the valorisation of non-marketable vegetables for the development of new value-added food products, within a framework of sustainability and the promotion of circular economy principles.

Low-caliber beetroot (*Beta vulgaris*) samples were sanitized and subjected to different pre-treatments using PEF (1.5 kV/cm; 5.4 kJ/kg) and HPP (450 MPa for 5 min) to enhance flavour incorporation by inducing cellular structural changes that modify membrane permeability and facilitate mass transfer. For the PEF pretreatment, beetroot samples were initially processed, then cut into cubes (10x10x10 mm) and mixed with salicornia and spices. For the HPP pretreatment, after sanitization, samples were cut into cubes (10x10x10 mm), mixed with salicornia and spices, vacuum-packaged, and subsequently subjected to HPP. Control samples were also prepared, consisting of cubed beetroot seasoned with salicornia and spices, without any pretreatment. Both pretreated and control samples were subsequently freeze-dried using an industrial freeze-dryer, with a total cycle time of 50 h and an initial temperature of −40 °C. Primary drying was carried out for 43 h, with temperatures ranging from −40 °C to 35 °C, under a vacuum between 1 mbar and 0.15 mbar. Secondary drying lasted 7 h, at temperatures between 37 °C and 40 °C, operating at an approximate vacuum of 0.025 mbar. Samples were then vacuum packed in polyamide/polyethylene bags to avoid rehydration. Sensory attributes (colour, aroma, flavour, texture, and overall acceptance) and purchase intention were evaluated. Analyses were performed in triplicate to evaluate physicochemical properties (texture and colour) and microbiological compliance within the scope of hygienic-sanitary control.

At the sensory level, both PEF and HPP pretreatments showed a positive impact compared to the control samples. However, between both technologies studied, HPP proved to be slightly more promising, particularly in colour and flavour. Samples treated with HPP were preferred by consumers and showed the highest purchase intention.

Regarding texture, significant differences were observed in the hardness parameter between control and those treated with PEF and HPP, indicating that the pretreatments influenced the mechanical resistance of the product. This result suggests increased crispness, in agreement with the sensory analysis findings.

When it comes to instrumental colour, significant differences were observed in the lightness (L*), greenness/redness (a*), and blueness/yellowness (b*) parameters in HPP-treated samples compared to the others (PEF and control), corroborating the sensory analysis results, particularly in terms of consumer preference for colour.

From a microbiological perspective, HPP treatment showed a significant effect in reducing microbial load when compared to the control and PEF pre-treated samples. Indeed, a 2-log reduction was observed in the total number of viable microorganisms at 30 °C, while *Enterobacteriaceae* counts were below the detection limit in HPP-treated samples, while no significant reduction in microbial load was observed for the PEF pre-treatment. These results highlight the potential of HPP, as a nonthermal technology, to enhance the microbiological safety of food products.

The results highlight the potential of PEF and HPP technologies, when applied as pretreatments, in the development of dehydrated products, with particular emphasis on the improvement of sensory properties, where HPP stood out compared to PEF. In addition, HPP demonstrated high efficacy in reducing microbial load, reinforcing its relevance as a nonthermal technology for the development of freeze-dried products. Overall, these findings underline the contribution of these approaches to the valorisation of low-commercial-value raw materials through the production of value-added dehydrated foods.

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O7: Effects of two pressure-based pasteurizations and packaging atmosphere on lipid oxidation of smoked salmon

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Lipid oxidation is one of the primary deterioration pathways in fatty fish products, compromising sensory quality, nutritional value, and shelf life[1]. Pressure-based technologies such as high-pressure processing (HPP) and moderate-pressure pasteurization (MPP) are increasingly explored as non-thermal preservation strategies[2,3], yet their influence on lipid oxidation dynamics in smoked fish, particularly in interaction with packaging atmosphere, remains poorly understood.
This study aimed to evaluate and compare the effects of HPP (600 MPa, 5 min) and MPP (225 MPa, 3.5 days), combined with three packaging atmospheres — atmospheric air, oxygen-scavenger, and 70% vacuum — on lipid oxidation progression in smoked sockeye salmon over 30 days of refrigerated storage (5 °C).
Lipid oxidation was monitored through a comprehensive set of chemical and instrumental indicators, including peroxide value (PV), thiobarbituric acid reactive substances (TBARS), fluorescence ratio, and volatile oxidation-related compounds (aldehydes, ketones, and hydrocarbons) identified and quantified throughout storage.
Both pressure treatments accelerated lipid oxidation relative to untreated controls, with MPP inducing more pronounced effects due to its extended duration. Elevated PV, TBARS, fluorescence ratio, and volatile aldehyde accumulation were observed in all pressure-treated samples, particularly those stored under air packaging. The combination of MPP with atmospheric air packaging represented the most oxidatively unstable condition across all measured parameters. Oxygen-scavenger packaging effectively suppressed lipid degradation in both HPP- and MPP-treated samples, maintaining oxidation markers at levels comparable to or below untreated controls. Vacuum packaging offered intermediate protection, while air packaging consistently yielded the highest degree of oxidative deterioration.
Packaging atmosphere emerged as the dominant factor governing lipid oxidation outcomes in pressure-treated smoked salmon. The pairing of pressure-based pasteurization with oxygen-scavenger packaging represents a viable strategy for simultaneously achieving microbial safety and oxidative stability. Conversely, the use of MPP without active oxygen management should be avoided. These findings provide critical guidance for the design of effective and commercially feasible processing and packaging strategies for smoked salmon products.

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O8: Changes in Quality-Related Compounds in Purple Carrot Juice after High-Pressure and Thermal Sterilization Treatments by PESI-MS/MS Analysis

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Purple carrot contains various plant-derived compounds, including acylated anthocyanins, and has attracted attention in the food processing field because of its colour stability and antioxidant properties. However, the effects of processing steps such as juicing, high pressure processing (HPP), and thermal sterilization on the molecular profiles of purple carrot components remain insufficiently understood.

In this study, purple carrot samples produced in Hokkaido, Japan, including intact raw material before juicing, freshly squeezed juice, HPP-treated juice, and thermally sterilized juice, were comparatively analysed using Probe Electrospray Ionization-Tandem Mass Spectrometry (PESI-MS/MS)[1]. HPP treatment was mainly conducted at 600 MPa for 3 min, and the differences in component profiles between non-thermal and thermal processing conditions were evaluated.

PESI-MS/MS analysis revealed differences in mass spectral patterns among the processing conditions. In particular, differences in peak intensity and distribution were observed not only for signals presumed to be associated with anthocyanins, but also for signals potentially derived from organic acids, sugars, amino acids, and polyphenolics. Both thermal treatment and HPP treatment exhibited distinct spectral profiles, suggesting that differences in processing methods may influence the molecular states of purple carrot-derived components.

These results suggest that PESI-MS/MS may be a useful rapid analytical approach for evaluating changes in plant-derived component profiles associated with food processing. Future studies will focus on detailed compound identification using HPLC-MS/MS and additional analyses of polymerization and intermolecular association states of purple carrot-derived polyphenols. Furthermore, potential relationships between these molecular state changes and interactions with G-quadruplex (G4) structures[2] will be investigated in future studies.

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O9: Effect of HHP treatment for the preservation of the olive pomace of the Morisca cultivar as a food additive

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Olive pomace is one of the main seasonal by-products from the olive oil industry, and a rich source of phenolic compounds with high antioxidant properties. Its adequate treatment can offer them a feasible alternative as a natural additive for food products with a high added value. The use of High Hydrostatic Pressures (HHP) as a nonthermal preservation technique can effectively control the microbiological load of this matrix during long-term refrigeration, while minimally impacting its active compounds and properties. Additionally, the HHP may inhibit the polyphenol oxidase (PPO) enzymes that are primarily responsible for degrading the phenolic compounds found in olive pomaces. The present study assessed the effect of the application of the HHP (600 MPa/5 minutes) on the microbiological load (mesophiles, psychrophiles, moulds and yeast), the inactivation of PPO, the contents of total phenolic compounds (TP) and total antioxidant activity (TAA) of the olive pomaces from the Morisca cultivar at two ripening stages (maturity index equal to 0.30 and 4.96, for green and fully ripe, respectively), stored for 4 months under refrigeration (4 °C) (T0: initial time. T1: two weeks, T2: one month; T3: four months). Samples with no HHP treatment were used as controls.

According to **Table 1**, at T0, the microbiological loads in all pomaces were low, with values below 2.5 log CFU g^{-1} for all microorganisms. While HHP significantly reduced loads of mesophiles in green samples, its critical preservative effect was observed in mature samples during storage. By T3, the total mesophiles increased in the mature control batch, reaching 4.57 log CFU g^{-1}, whereas HHP treatment reduced this to 1.12 log CFU g^{-1}. The total counts of moulds and yeast significantly increased during storage, particularly in the mature control samples, which reached 4.79 log CFU g^{-1}, whereas the HHP-treated samples maintained counts at 1.41 log CFU g^{-1}. For psychrophiles, an increase was observed only in the treated green pomaces at T3, reaching values close to the detection limit of 1.18 log CFU g^{-1}.

Table 1. Microbiological loads of the olive pomaces treated with HHP and evolution during storage (log CFU g^{-1})

		Morisca green		Morisca mature		
		Control	600 MPa	Control	600 MPa	Sig.
Total mesophiles	T0	1.95 ± 0.15 a AB	1.24 ± 0.27 b	2.12 ± 0.26 a B	1.61 ± 0.25 ab	***
	T1	1.82 ± 0.24 ab AB	1.12 ± 0.24 b	2.08 ± 0.18 a B	1.15± 0.96 b	**
	T2	3.35 ± 1.7 A	2.23 ± 1.99	3.65 ± 1.26 AB	2.09 ± 1.67	ns
	T3	nd b B	nd b	4.57 ± 0.04 a A	1.12 ± 0.24 b	***
	Sig.	**	ns	**	ns	
Total moulds and yeasts	T0	nd b B	nd b B	1.65 ± 0.66 a C	1.17 ± 0.29a	***
	T1	1.16 ± 0.45 AB	1.13 ± 0.39 A	1.88 ± 0.19 C	1.03 ± 0.13	ns
	T2	1.13 ± 0.23 b AB	nd c B	3.44 ± 0.48 a B	0.96 ± 0.10 b	***
	T3	1.48 ± 0.51 b A	1.11 ± 0.23 b A	4.79 ± 0.24 a A	1.41 ± 0.46 b	***
	Sig.	**	***	***	ns	
Total psychrophiles	T0	nd b B	nd b B	1.28 ± 0.34 a A	0.96 ± 0.11 a A	***
	T1	nd B	nd B	nd B	nd B	ns
	T2	1.05 ± 0.27 a A	nd b B	nd b B	nd b B	***
	T3	nd b B	1.18 ± 0.34 a A	nd b B	nd b B	***
	Sig.	***	***	***	***	

Small letters in the same row indicate significant differences among pomaces. Capital letters in the same column indicate significant differences among storage times. nd: not detected, and limit of detection= 1 log CFU g^{-1}. CFU: colony-forming unit. Sig.: Significance. ANOVA test significance levels: ns p>0.05; * p≤ 0.05; ** p≤ 0.01; *** p≤ 0.001.

The highest PPO activity was associated with mature samples, while the green samples showed minimal activity (**Figure 1**). For the mature batches, the HHP resulted in a decrease in PPO activity during storage, although this change was not statistically significant.

For TAA and TP (**Table 2**), initially (T0), the treated green samples showed the highest TAA and the treated mature samples the lowest, showing the control batches intermediate values. At T3, a significant decrease of TAA took place in all the batches, with the maximum values linked to control green pomaces and the minimum

values to the treated mature pomaces, and the application of HHP had no significant effect. The TP contents at T0 and T1 were similar in all the batches. At T3, only a significant decrease was observed in the mature treated pomaces, showing the lowest values of TP.

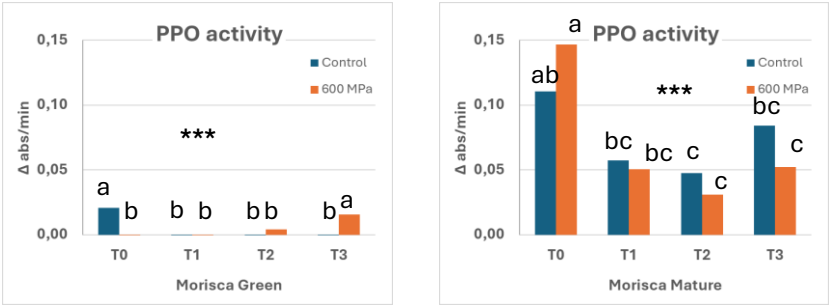

Figure 1. Polyphenol oxidase (PPO) activity of olive pomaces throughout storage for each olive type. a, b, c: different letters indicate significant differences between pomaces within a type of olive. Significance levels: *** p≤ 0.001.

Table 2. Total antioxidant activity (TAA), total phenols content (TP), of the olive pomaces treated with HHP and evolution during storage.

		Morisca green		Morisca mature		
		Control	**600 MPa**	**Control**	**600 MPa**	**Sig.**
TAA	T0	70.19 ± 0.43 ab A	73.01 ± 2.65 a A	66.45 ± 3.00 ab A	62.43 ± 5.62 b A	**
	T1	46.70 ± 3.61 B	49.75 ± 2.41 B	50.78 ± 2.21 B	53.68 ± 4.46 A	ns
	T2	50.76 ± 2.28 a B	47.07 ± 4.79 ab B	39.98 ± 4.75 c C	38.20 ± 3.07 c B	***
	T3	48.99 ± 3.86 a B	43.39 ± 5.03 ab B	39.36 ± 4.59 ab C	33.05 ± 3.44 b B	***
	Sig.	***	***	***	***	
TP	T0	5.91± 0.16 A	5.73± 0.06 A	5.53± 0.45 A	5.26± 0.27 A	ns
	T1	4.59± 0.23 B	4.35± 0.17 BC	4.48 ± 0.23 AB	4.79 ± 0.40 A	ns
	T2	4.78 ± 0.28 a AB	4.29 ± 0.54 ab C	3.86 ± 0.30 bc B	3.56 ± 0.88 c B	***
	T3	5.20 ± 0.66 a AB	4.77 ± 0.30 ab AB	4.35 ± 0.58 ab AB	3.65 ± 0.49 c B	***
	Sig.	*	***	**	***	

TAA: total antioxidant activity (µmol Trolox equivalent g-1 pomace). TP: total content of phenols (mg of gallic acid equivalent g-1 pomace). Small letters in the same row indicate significant differences among pomaces. Capital letters in the same column indicate significant differences among storage times Sig: significance of ANOVA test. Significance levels: ns p>0.05; * p≤ 0.05; ** p≤ 0.01; *** p≤ 0.001.

This study suggests that HHP treatment at 600 MPa for 5 minutes can effectively reduce the microbiological load of Morisca mature olive pomaces, facilitating their long-term preservation in refrigeration. However, for green pomaces, since untreated control samples naturally maintained low microbial counts throughout the storage period, HHP treatment proved to be less critical for their preservation. Additionally, the treatment did not negatively alter the levels of total phenolic (TP) compounds and total antioxidant activity (TAA), and it was effective in inactivating polyphenol oxidase (PPO), particularly in the mature pomaces.

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O10: Effects of sodium chloride and osmotic conditions on the barotolerance of *Staphylococcus aureus* under high hydrostatic pressure treatment

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Staphylococcus aureus is a foodborne pathogen known for its high resistance to environmental stresses, including salt and pressure[1-3]. Although this bacterium is non-spore-forming, it has been reported to survive even after high hydrostatic pressure (HHP) treatment at 600 MPa, a pressure is commercially used worldwide[3]. However, its pressure resistance mechanism remains unclear. Additionally, our previous studies have suggested that the bactericidal effect of HHP treatment may be influenced by food matrices, such as food components and osmotic pressure[4]. The objective of this study is to clarify whether factors such as sodium chloride (NaCl) and osmotic pressure are involved in the barotolerance of *S. aureus* and the bactericidal effects of HHP treatment. We examined the change in the bactericidal effects of HHP treatment on *S. aureus* when NaCl and other solutes were added and then adjusted the osmotic pressure of the broth.

First, the effect of NaCl addition before and after HHP treatment on the bactericidal effectiveness against *S. aureus* was assessed. A culture of *S. aureus* NBRC12732 was inoculated into brain heart infusion (BHI) broth at approximately 6–7 log CFU/mL, followed by HHP treatment (400 MPa, 25°C, 10–30 min). NaCl (0–21%) was added to the broth either before or after HHP treatment. After HHP treatment, samples were incubated at 37°C for 24 h. Bacterial counts were determined using the plate count method on BHI agar. When NaCl was added before HHP treatment, the bacterial counts immediately after HHP treatment were higher than in samples without NaCl during HHP treatment. However, when the effect of the timing of NaCl addition on the bactericidal efficacy of HHP treatment was examined (**Figure 1**), no significant difference in bacterial counts was observed between pre- and post-treatment addition at 7% NaCl. In contrast, at higher concentrations (≥15%), bacterial counts were lower when NaCl was added after HHP than when added beforehand. These findings suggest that NaCl presence during HHP provides a protective effect for *S. aureus*, thereby reducing the bactericidal effectiveness of the treatment.

Next, we compared the bactericidal effects of HHP treatment on *S. aureus* in a liquid medium adjusted to a consistent osmolality level using various solutes. A modified minimal medium for *S. aureus* was prepared by respectively adjusting the concentrations of NaCl, potassium chloride (KCl), calcium chloride ($CaCl_2$), and glucose to set an osmolarity of approximately 2500 mOsm/kgH_2O. Each medium was inoculated with approximately 9 log CFU/mL of *S. aureus* and subjected to HHP treatment (600 MPa, room temperature, 10 min). Compared to the control group without added solutes, the bacterial count immediately after HHP treatment was higher in all media containing added solutes, regardless of the solute type. These results indicate that the reduced bactericidal efficacy is not specific to NaCl, but rather that high osmotic pressure itself diminishes the effectiveness of HHP treatment.

Finally, we examined the effect of food osmotic pressure on the bactericidal efficacy of HHP treatment against *S. aureus* using commercially available foods. We inoculated relatively high-osmotic-pressure liquid foods, such as syrups and dressings, with *S. aureus*, subjected them to HHP treatment, and then measured bacterial counts. Although some high-osmolarity foods, like chocolate sauce, honey, and sweetened adzuki beans, tended to show higher bacterial survival rates after treatment compared to low-osmolarity foods such as water and oolong tea, no correlation was observed between the food's osmolarity and the bacterial count after treatment.

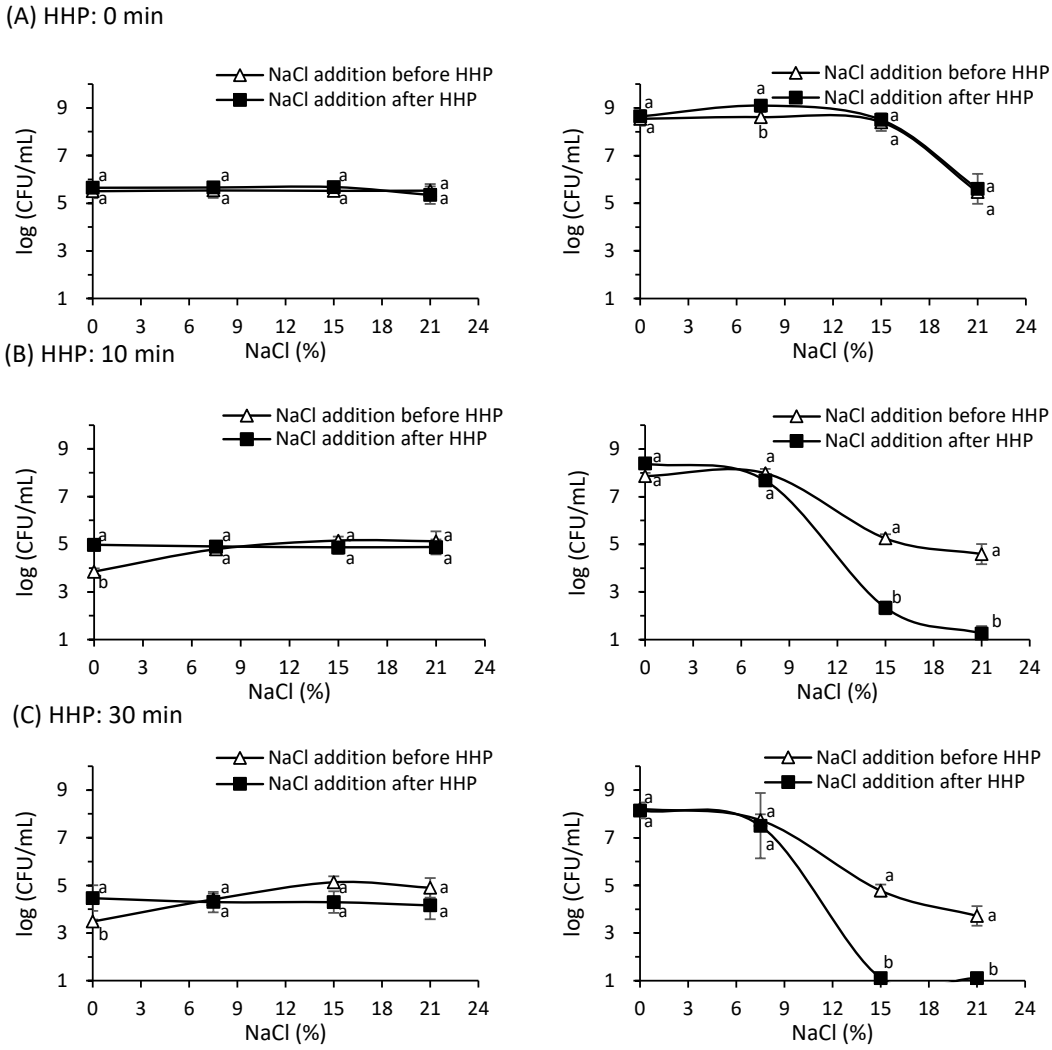

Figure 1. Effect of NaCl addition timing during HHP treatment on *S. aureus* NBRC 12732 before (left) and after 24 h of incubation (right). The detection limits of bacterial number were about 1 log CFU/mL. Values within the same column followed by different letters are significantly different ($p < 0.05$; $n = 3$).

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P1: Application of High Pressure for Yogurt Preservation

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Thermal pasteurization (TP) and refrigeration are conventional methods for food pasteurization and preservation. However, growing consumer demand for minimally processed, high-quality, and safe foods has driven interest in non-thermal technologies. High-pressure processing (HPP) is a well-established non-thermal pasteurization method operating at 400–600 MPa for a few minutes based on isostatic and *Le Chatelier's* principles [1, 2]. Because TP compromises yogurt quality and HPP may fail to inactivate lactic acid bacteria (LAB) or can trigger spore germination, hyperbaric storage (HS) at room temperature (RT) using moderate pressures has emerged as a promising alternative for simultaneous pasteurization and preservation [3, 4, 5]. This study evaluated HS at RT (19–22 °C) for commercial natural yogurt, testing 175 MPa for pasteurization during storage and 75 MPa as an energy-saving alternative to traditional refrigeration (RF, 4 °C). Spoilage microorganisms (total psychrotrophiles, Enterobacteriaceae, yeasts, and molds) and LAB viability were monitored over 168 hours. Physicochemical properties (pH, titratable acidity [TA], total soluble solids [◦Brix], and CIELAB color), syneresis, and water holding capacity were evaluated up to 504 hours.

Spoilage microorganisms remained below the detection limit (2.70 log CFU/g) across all treatments. While RF preserved LAB viability, HS inactivated LAB by 120 hours at 75 MPa and within 3 hours at 175 MPa. Physicochemically, ◦Brix and color remained highly stable (ΔE^* <1.52) under all conditions. RF maintained stable TA (~10.88 g/L lactic acid) and pH (4.10), while syneresis gradually increased to 53.62%. Excluding a potential 48-hour anomaly, the 75 MPa treatment demonstrated overall physicochemical stability, with pH ranging from 3.86–4.16, TA stabilizing at 10.10–10.30, and syneresis recovering to 50.45% by 168 hours. In contrast, HS at 175 MPa rapidly inactivated LAB within 3 hours, preventing post-acidification due to LAB metabolism and maintaining a more stable pH (4.11–4.15), TA (8.07–9.87), and syneresis profile (51.64–56.09%).

In conclusion, HS at RT, particularly at 175 MPa, successfully preserved yogurt quality. While LAB inactivation strips the yogurt of its conventional "live-culture" status, it prevents over-acidification, offering a distinct advantage for ambient-temperature shelf-life extension. Future research should explore pressures below 75 MPa to preserve LAB viability and maintain the legal classification of yogurt while exploiting the energy-saving benefits of HS.

Acknowledgements and funding

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P2: High Hydrostatic Pressure Treatment on White Winemaking Lees (cv. Alarije)

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White winemaking lees (cv. Alarije) are an oenological by-product generated after alcoholic fermentation as a consequence of the sedimentation of yeasts, cellular debris, and other compounds derived from must and wine. These lees exhibit a heterogeneous composition rich in polysaccharides, proteins, and phenolic compounds, which has attracted increasing interest regarding their characterization and potential valorization. However, as a seasonal by-product generated only during the harvest season, strategies to improve its preservation and long-term valorization are needed. In this context, high hydrostatic pressure (HHP) has emerged as a promising technology to enhance microbiological stability while preserving bioactive compounds. Therefore, this study evaluated the effect of different HHP treatments on the microbiological stability and phenolic content of winemaking lees stored under refrigerated conditions. After one month of natural settling at room temperature, the solid (S) and liquid (L) fractions were separated and divided into an untreated control and three HHP treatments. HHP1 (400 MPa, 1 min), HHP2 (500 MPa, 1 min), and HHP3 (600 MPa, 1 min) in a commercial Hiperbaric unit (Wave 6000/55) with an initial water temperature of 10ºC. Subsequently, all samples were stored at 4 °C in order to evaluate the changes during the refrigerated storage (90 days).

Microbiological counts, expressed as log CFU (colony forming units) (**Table 1**), showed significant differences between the solid and liquid fractions with highest counts in the solid fractions. Mesophilic microorganism counts in the solid fraction were not affected after HHP treatment in both fractions at day 1. However, after 90 days of storage, a reduction in mesophilic counts was observed in HHP-treated samples compared to the untreated controls (S and L), particularly at higher pressure levels (HHP3), while HHP1 and HHP2 showed intermediate values. Regarding molds and yeasts, in the solid fractions HHP treatments did not reduce the counts at day 1, however, at day 90, HHP2 and HHP3 showed lower counts than HHP1 and control. Therefore, these more intense processing conditions were more effective to maintain the counts of mesophilic and molds and yeast at low levels. In the liquid fraction the levels of molds and yeast was below the detection limit after HHP. These findings are consistent with previous studies describing the ability of HHP to induce sublethal damage in microorganisms and limit their growth during storage[1,2]. In other winery by-products like pomace, HHP was effective in achieving microbiological stability without the need for intensive thermal treatments [1,3] .

Table 1. Microbiological count (log UFC g^{-1}) during storage (90 days) at refrigerated temperatures after high pressure treatment of white wine lees.

White Wine Lees										
		Control		HHP 1		HHP 2		HHP 3		
		Solid	Liquid	Solid	Liquid	Solid	Liquid	Solid	Liquid	*p-value*
		Mesophilics counts								
Storage (days)	1	4.6a ± 0.1	2.3b ± 0.3	4.5a ± 0.1	2.4b ± 0.1	4.5a ± 0.0	2.4b ± 0.2	4.4a ± 0.1	2.3b ± 0.2	***
	90	4.5a ± 0.2	2.5c ± 0.0	4.4ab ± 0.0	2.3c ± 0.1	4.3ab ± 0.1	2.2c ± 0.0	4.2b ± 0.2	2.2c ± 0.1	***
p-storage		ns	ns	ns	ns	*	ns	ns	ns	
		Molds and yeast								
Storage (days)	1	2.3a ± 0.1	<1.0b	2.2a ± 0.1	<1.0b	2.2a ± 0.4	1.3b ± 0.5	2.2a ± 0.3	<1.0b	***
	90	2.0a ± 0.2	<1.0b	1.8a ± 0.1	1.0b ± 0.0	1.2b ± 0.3	1.1b ± 0.2	1.1b ± 0.0	<1.0b	***
p-storage		ns	ns	ns	ns	ns	ns	ns	ns	

Results are expressed as mean ± standard deviation and significance (p) from a one-way ANOVA. Different letters in the same row indicates significant differences * (p<0.05), ** (p<0.01) and *** (p<0.001) between different samples in Tukey's test. Control (No HHP), HHP1 (400 MPa, 1 min), HHP2 (500 MPa, 1 min) and HHP3 (600 MPa, 1 min).

Regardless of treatment, total phenolic compounds (**Table 2**) were significantly higher in the solid fractions than in the liquid fractions ($p < 0.001$), confirming the greater capacity of solid lees to retain phenolic compounds. HHP treatments largely preserved phenolic content in both fractions immediately after processing. HHP preserved phenolic compounds after HHP in both fractions, although refrigerated storage led to an overall decrease in phenolic content. After 90 days of storage, solid fraction treated by HHP presented lower levels than the non-treated samples, while liquid fraction presented similar levels treated and non-treated.

Thus, the preservation of phenolics of solid fractions of lees during storage should be improved. HHP-treated samples maintained relevant levels of these compounds after 90 days of storage. This reduction could be related to oxidative degradation phenomena, as previously described in wines and winery by-products treated with HHP. This effect could be related to enhanced matrix stabilization and reduced oxidative degradation, as previously described in wines and winery by-products treated with high hydrostatic pressures [2,4].

Table 2. Total phenolic compounds content (mg GAE 100 g^{-1}) during storage (90 days) at refrigerated temperatures after high pressure treatment of white wine lees.

White Wine Lees										
		Control		HHP 1		HHP 2		HHP 3		
		Solid	Liquid	Solid	Liquid	Solid	Liquid	Solid	Liquid	*p-value*
Storage (days)	1	133.3 a± 1.7	54.2b ± 5.5	143.3a ± 2.5	63.0b ± 2.1	138.6a ± 0.7	62.2b ± 4.1	139.3a ± 7.3	41.2c ± 3.7	***
	90	106.6a ± 3.7	40.9c ± 1.8	86.7b ± 4.1	47.67bc ± 3.0	80.7b ± 3.2	55.8c ± 0.7	77.5b ± 5.7	52.9c ± 7.1	***
p-storage		***	*	***	**	***	ns	***	ns	

Results are expressed as mean ± standard deviation and significance (p) from a one-way ANOVA. Different letters in the same row indicates significant differences * (p<0.05), ** (p<0.01) and *** (p<0.001) between different samples in Tukey's test. Control (No HHP), HHP1 (400 MPa, 1 min), HHP2 (500 MPa, 1 min) and HHP3 (600 MPa, 1 min).

Overall, the results obtained suggest that HHP is a promising technological strategy for the valorization of winemaking lees. This treatment improves the microbiological stability of the by-product, although the stability of phenolic compounds during storage should be further enhanced, likely by reducing the activity of oxidative enzymes such as polyphenol oxidase. Nevertheless, HHP offers an attractive approach for the integral valorization of this by-product, as it avoids the use of solvents and enables the complete utilization of the entire material.

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P3: Effect of High Hydrostatic Pressures on Red Winemaking Lees (cv. Tempranillo)

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End-of-alcoholic-fermentation lees from red winemaking (*Vitis vinifera* L. cv. Tempranillo) constitute an oenological by-product resulting from the sedimentation of yeast biomass, cellular fragments of grape origin, and associated colloidal and precipitated components during alcoholic fermentation, and are characterized by a high content of bioactive compounds, particularly phenolic compounds. Due to their composition, these by-products have attracted increasing attention as potential sources of value-added- ingredients for food and biotechnological applications. This work aimed to investigate the effects of high hydrostatic pressure (HHP) treatments on the microbiological quality and phenolic composition of red winemaking lees during refrigerated storage. After one month of natural settling at room temperature, the lees were separated into solid and liquid fractions (**Figure 1**). Each fraction was subsequently divided into four experimental groups: an untreated control and three HHP treatments (HHP1: 400 MPa for 1 min; HHP2: 500 MPa for 1 min; HHP3: 600 MPa for 1 min), applied using a commercial Hiperbaric unit (Wave 6000/55) with an initial water temperature of 10 °C. All samples were then stored at 4 °C for 90 days to evaluate changes during refrigerated storage.

Figure 1. Natural decanting separation of the different red wine lees fractions.

Microbiological analysis (**Table 1**) revealed marked differences between solid and liquid fractions. Microorganism counts were generally higher in the solid fraction than in the liquid fraction. For mesophilic microorganisms HHP reduced counts relative to the untreated control at day 1 with the effect more evident in the liquid fractions. After 90 days of refrigerated storage, mesophilic counts showed comparable values among treated and untreated samples both liquid and solid fractions. A similar trend to observed for mesophilic populations was found for molds and yeast. The highest counts were found in the solid fraction, and they were reduced by HHP treatments. In the liquid fraction, counts were generally at or below the detection limits whit the exception of the HHP3 samples on day 1. This slight increase of counts in liquid fraction could be caused because HHP may induce physiological changes in fungal spores that favor germination or metabolic activation after treatment, especially when sublethal pressures are applied [1]. These observations are in agreement with previous studies reporting the effectiveness of HHP treatments in reducing microbial viability and improving the microbiological stability of winery by-products[1,2]. Comparable results have been reported in grape-derived matrices processed by HHP, where microbial stabilization was achieved without the application of severe thermal treatments[1].Furthermore, higher pressures levels showed greater antimicrobial efficacy, indicating an increased sensitivity of spoilage microorganisms to HHP treatments.

Table 1. Microbiological count (log UFC g^{-1}) during storage (90 days) at refrigerated temperatures after HHP of red wine lees.

Red Wine Lees										
		Control		HHP 1		HHP 2		HHP 3		
		Solid	Liquid	Solid	Liquid	Solid	Liquid	Solid	Liquid	*p-value*
		Mesophilics counts								
Storage (days)	1	4.6a ± 0.1	3.2b ± 1.1	3.4b ± 0.4	1.4c ± 0.3	3.1 b± 0.0	1.6c ± 0.0	3.3b ± 0.3	1.7c ± 0.2	***
	90	3.7a ± 0.0	1.7b ± 0.2	3.4a ± 0.2	1.3b ± 0.3	3.4a ± 0.1	1.8b ± 0.2	3.5a ± 0.1	1.5b ± 0.4	***
p-storage		***	ns	ns	ns	**	ns	ns	ns	
		Molds and yeast								
Storage (days)	1	1.8a ± 0.0	<1.0	1.2b ± 0.2	1.0b ± 0.0	1.1b ± 0.2	<1.0	<1.0	1.2b ± 0.4	***
	90	1.7a ± 0.2	<1.0	1.5a ± 0.1	1.0b ± 0.0	1.2b ± 0.2	<1.0	1.1b ± 0.2	<1.0	***
p-storage		ns	ns	ns	ns	ns	ns	ns	ns	

Results are expressed as mean ± standard deviation and significance (p) from a one-way ANOVA. Different letters in the same row indicate significant differences * (p<0.05), ** (p<0.01) and *** (p<0.001) between different samples in Tukey's test. Control (No HHP), HHP1 (400 MPa, 1 min), HHP2 (500 MPa, 1 min) and HHP3 (600 MPa, 1 min).

Concerning total phenolic compounds (**Table 2**), the solid fraction consistently showed significantly higher concentrations than the liquid fraction ($p < 0.001$), confirming the strong association of phenolic compounds with the solid matrix of red lees. Although a general decline in phenolic content was observed during refrigerated storage, HHP-treated samples retained considerable levels of phenolic compounds after 90 days of storage. In some treated liquid phenolic contents at day 90 were higher than those measured the beginning of storage. This increase may be attributed to pressure-induced disruption of cellular structures, which can facilitate the release of bound phenolic compounds [1,3].

Table 2. Total phenolic compounds content (mg GAE 100 g^{-1}) during storage (90 days) at refrigerated temperatures after high pressure treatment of red wine lees.

Red Wine Lees										
		Control		HHP 1		HHP 2		HHP 3		
		Solid	Liquid	Solid	Liquid	Solid	Liquid	Solid	Liquid	*p-value*
Storage (days)	1	280.6a ± 9.6	121.5d ± 22.5	255.5ab ± 9.5	208.7bc ± 7.8	241.3ab ± 10.8	217.5bc ± 54.3	264.3ab ± 16.1	162.3cd ± 13.3	***
	90	275.2a ± 4.6	186.4c ± 23.1	231.0b ± 9.6	213.5bc ± 19.8	220.4bc ± 15.8	180.0c ± 11.9	189.5bc ± 11.5	230.4b ± 20.5	***
p-storage		ns	*	*	ns	ns	ns	**	*	

Results are expressed as mean ± standard deviation and significance (P) from a one-way ANOVA. Different letters in the same row indicates significant differences * (P<0.05). ** (P<0.01) *** (P<0.001) between different treatments in Tukey's test. Control (No HHP). HHP1 (400 MPa, 1 min). HHP2 (500 MPa, 1 min). HHP3 (600 MPa, 1 min).

Overall, these findings demonstrate that HHP processing may represent an effective strategy for extending the stability and enhancing the valorization potential of red winemaking lees, while preserving relevant amounts of phenolic compounds during refrigerated storage.

Acknowledgements and funding

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P4: High Hydrostatic Pressure (HHP) as a Preservation Strategy for Frankfurters Enriched with Grape Pomace

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The development of meat products with reduced or no synthetic additives represents an important challenge for the industry due to their lower oxidative and microbiological stability and the consequent reduction in shelf-life. In this context, High Hydrostatic Pressure (HHP) has gained increasing attention as an effective non-thermal preservation technology, capable of extending product shelf-life through microbial inactivation while minimizing changes in nutritional and sensory quality[1]. The combination of HHP with natural ingredients rich in bioactive compounds may further enhance product stability and provide an alternative strategy for the development of clean-label meat products.

Winemaking by-products such as grape pomace still contain significant amounts of phenolic compounds and other functional constituents with recognized antioxidant and antimicrobial activity, making them promising ingredients for food preservation applications[2,3]. Their incorporation into meat systems, together with HHP processing, could contribute to improving oxidative stability and limiting microbial growth, particularly in formulations without conventional additives.

Therefore, this study investigated the combined application of High Hydrostatic Pressure (HHP) and bioactive ingredients derived from red and white grape pomace as a preservation strategy to improve the shelf-life of additive-free pork frankfurters. HHP treatment was performed using a Hiperbaric 6000/55 equipment under processing conditions of 600 MPa for 3 min, with an initial temperature of 10 °C.

The results obtained from the combined application of High Hydrostatic Pressure (HHP) and the incorporation of grape pomace-derived bioactive ingredients in pork frankfurter formulation are presented in **Figures 1** and **2**. **Figure 1** shows the results of microbiological results, including mesophilic counts, psychrophilic counts, and moulds and yeasts counts, evaluated immediately after processing and at the end of storage. Figure 2 presents the lipid oxidation values, illustrating the effect of both preservation strategies on oxidative stability during storage.

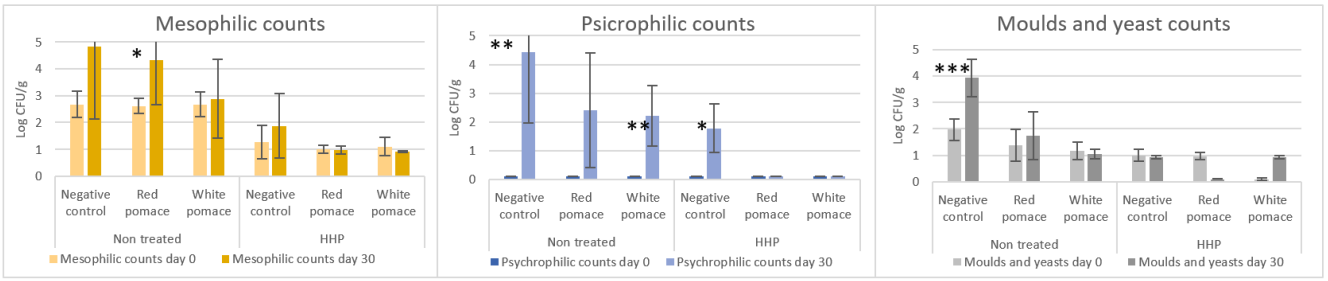

Figure 1. Microbiological counts of pork frankfurters formulated as negative control (without additives) and with bioactive ingredients obtained from red and white grape pomace, under non-treated and High Hydrostatic Pressure (HHP)-treated conditions. Measurements were performed at day 0 and day 30 of storage to assess the effect of HHP and grape-derived ingredients on product stability.

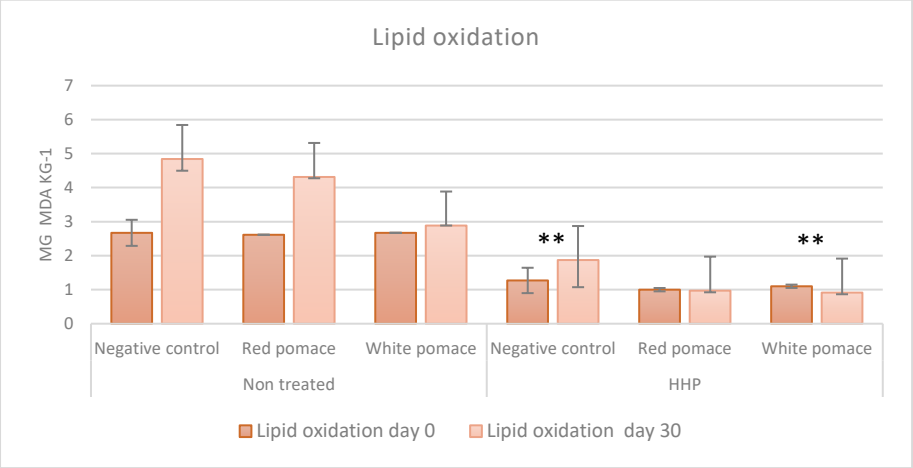

Figure 2. Lipid oxidation (mg MDA kg $^{-1}$) of pork frankfurters formulated as negative control (without additives) and with bioactive ingredients obtained from red and white grape pomace, under non-treated and High Hydrostatic Pressure (HHP)-treated conditions. Measurements were performed at day 0 and day 30 of storage to assess the effect of HHP and grape-derived ingredients on product stability.

High Hydrostatic Pressure (HHP) treatment showed a clear positive effect on the preservation of pork frankfurters, mainly through the reduction of microbial growth and the improvement of oxidative stability during storage. In general, HHP-treated samples exhibited lower microbiological counts than non-treated samples, indicating the effectiveness of pressure processing in controlling spoilage microorganisms and extending product shelf-life. In addition, HHP contributed to reducing lipid oxidation levels, resulting in improved oxidative stability and preservation of product quality.

The incorporation of bioactive ingredients obtained from red and white grape pomace provided additional preservation benefits. Compared with the negative control, pomace-containing formulations showed lower microbial counts at the end of storage, particularly for psychrophilic microorganisms, supporting their antimicrobial potential. Furthermore, grape pomace ingredients improved oxidative stability by reducing lipid oxidation values, which exhibited the strongest antioxidant effect.

Overall, the combined application of HHP and grape pomace ingredients demonstrated a complementary effect, improving the microbiological and oxidative stability of additive-free frankfurters.

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P5: Application of high hydrostatic pressure technologies and skin packaging for the preservation of ready-to-eat lamb meat dishes

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Current food trends show increasing consumer demand for sustainable, locally sourced foods with high sensory quality, nutritional value, and minimal preparation requirements. In this context, lamb production represents an important environmental and socioeconomic resource for rural areas, although the consumption of sheep meat remains limited and often seasonal[1]. At the same time, the market for ready-to-eat (RTE) products has expanded considerably, driven by changing lifestyles and the preference for convenient foods.

The development of innovative lamb-based RTE meals may contribute to increasing the consumption of these products while adding value to sheep production systems. However, ensuring product safety and extending shelf life without compromising quality remains a challenge. In this regard, high hydrostatic pressure (HHP) processing has emerged as a promising non-thermal preservation technology, allowing microbial inactivation while maintaining sensory and nutritional properties[2]. Therefore, the aim of this study was to evaluate the application of HHP combined with vacuum skin packaging as a preservation strategy for lamb-based RTE dishes, improving their microbiological stability, sensory quality, and commercial shelf life.

For this purpose, a traditional lamb dish, "*caldereta de cordero*", was prepared, packaged in vacuum skin trays containing 350 g of product, and subjected to two different HHP treatments (400 and 600 MPa for 5 min) using a semi-industrial Hiperbaric Wave 6000/55 unit with a 55 L capacity. A non-treated batch served as the control treatment for comparison. All samples were subsequently stored under refrigeration for 100 days. Microbiological analyses were performed immediately after processing and at the end of storage in both HHP-treated and control samples to evaluate the effect of pressure treatment on microbial counts and their evolution during refrigerated storage. In addition, sensory evaluation was conducted after processing by a trained panel, which carried out a hedonic assessment of general appearance, odour, flavour, texture and overall evaluation to determine the impact of the treatments on product quality and consumer acceptability.

Following HHP treatment, microbiological analyses of the lamb "*caldereta"* showed that all microbial counts on day 1 (after processing and treatment) were below the detection limit. After 100 days of refrigerated storage, microbial levels remained within the established microbiological standards. Mesophilic counts were lower in HHP-treated samples compared with the control batch, while the remaining microorganisms were not detected at any sampling time (**Table 1**).

Table 1. Microbiological analysis (log colony-forming units: log CFU/g) of the lamb caldereta ready-to-eat dish before processing and after high hydrostatic pressure treatment.

Day 1	Mesophilic counts	Psychrotrophic counts	*E. coli*	*S. aureus*	*Clostridium*	*Salmonella*	*L. monocytogenes*
Control	<1	<1	<1	<1	<1	Abs.	Abs.
400MPa 5'	<1	<1	<1	<1	<1	Abs.	Abs.
600MPa 5'	<1	<1	<1	<1	<1	Abs.	Abs.
Day 100							
Control	1,9	<1	<1	<1	<1	Abs.	Abs.
400MPa 5'	1,3	<1	<1	<1	<1	Abs.	Abs.
600MPa 5'	<1	<1	<1	<1	<1	Abs.	Abs.

<1 log = <10 CFU/g (below the detection limit). Abs. = absence of pathogens in 25 g of sample.

Sensory evaluation also showed positive results, as panellists did not detect differences between non-treated and pressure-treated lamb "*caldereta"*, indicating that HHP processing did not negatively affect product sensory quality (**Figure 1**).

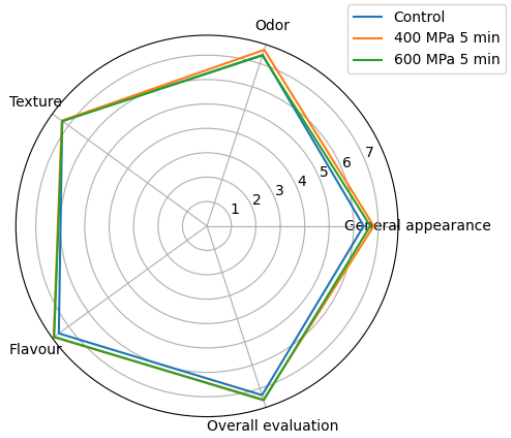

Figure 1. Results of the sensory evaluation of the ready-to-eat "*caldereta*" after preparation and treatment with HHP.

In conclusion, these results highlight the potential of high hydrostatic pressure (HHP) as an effective preservation technology for lamb-based ready-to-eat products, ensuring microbiological safety and extending shelf life while maintaining sensory quality without the need for additives. The combination of HHP and skin packaging may therefore represent a valuable strategy for the development of innovative ready-to-eat lamb dishes, facilitating the commercialization of traditional preparations such as lamb "*caldereta*". This approach could contribute to increasing consumer acceptance and consumption of lamb meat by adapting traditional products to current market trends focused on convenience, quality, food safety, and sustainable local production systems.

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P6: Effect of HHP treatment for the preservation of the olive pomace of the Picual cultivar as a food additive

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Olive pomace is one of the main seasonal by-products from the olive oil industry, and a rich source of phenolic compounds with high antioxidant properties. Its adequate treatment can provide a feasible alternative for its use as a natural, high-value food additive. The use of High Hydrostatic Pressures (HHP) as a nonthermal preservation technique can effectively control the microbiological load of this matrix during long-term refrigeration, while minimally impacting its active compounds and properties. Additionally, the HHP may inhibit the polyphenol oxidase (PPO) enzymes that are primarily responsible for degrading the phenolic compounds found in olive pomaces. The present study assessed the effect of the application of the HHP (600 MPa/5 minutes) on the microbiological load (mesophiles, psychrophiles, moulds and yeast), the inactivation of PPO, the contents of total phenolic compounds (TP) and total antioxidant activity (TAA) of the olive pomaces from the Picual cultivar at two ripening stages (mature index equal to 0.35-green and 3.12-fully ripe, respectively), stored for 4 months under refrigeration (4 °C) (T0: initial time. T1: two weeks, T2: one month; T3: four months). Samples with no HHP treatment were used as controls.

At the beginning of the assay, the pomaces exhibited low levels of microorganisms, with counts below 2.5 log CFU g^{-1}. The high hydrostatic pressure (HHP) treatment was only effective for the mature samples, significantly reducing their counts of mesophiles, moulds, yeasts, and psychrophiles. After storage, the control batches showed a general increase in mesophiles, moulds, yeasts, and psychrophiles, except for the green variety. The HHP treatment effectively controlled the microbiological loads, resulting in a significant reduction of mesophiles, moulds, yeasts, and psychrophiles at the end of the storage period (T3) (**Table 1**).

Table 1. Microbiological loads of the olive pomaces treated with HHP and evolution during storage (log CFU g^{-1}).

		Picual green		Picual mature		
		Control	600 MPa	Control	600 MPa	Sig.
Total mesophiles	T0	2.16 ± 0.11 a	2.08 ± 0.23 a A	2.38 ± 0.23 a B	1.28 ± 0.34 b A	***
	T1	2.80 ± 1.21 a	0.96 ± 0.11 b B	2.26 ± 0.06 ab B	0.96 ± 0.11 b A	**
	T2	2.17 ± 0.04 a	nd c B	2.07 ± 0.07 a B	1.05 ± 0.27 b A	***
	T3	4.13 ± 1.03 a	0.82 ± 0.24 b B	4.42 ± 0.43 a A	nd b B	***
	Sig.	ns	**	***	***	
Total moulds and yeasts	T0	2.01 ± 0.22 ab B	1.24 ± 0.59 b A	2.24 ± 0.15 a B	nd c B	***
	T1	2.80 ± 1.18 AB	1.45 ± 0.48 AB	2.32 ± 0.22 B	1.66 ± 0.36 A	ns
	T2	1.93 ± 0.17 ab B	nd c B	2.39 ± 0.48 a B	1.25 ± 0.61 b A	***
	T3	4.04 ± 0.96 a A	1.51 ± 0.53 b A	4.82 ± 0.15 a A	nd c B	***
	Sig.	*	*	***	***	
Total psychrophiles	T0	nd b	0.97 ± 0.12 ab A	2.09 ± 1.04 a AB	nd b	***
	T1	1.92 ± 1.77	nd B	1.37 ± 0.81 B	nd	ns
	T2	0.96 ± 0.11 b	nd c B	1.52 ± 0.14 a B	nd c	***
	T3	2.67 ± 1.50 ab	0.96 ± 0.11 ab A	3.18 ± 1.46 a A	nd b	***
	Sig.	ns	***	ns	ns	

Small letters in the same row indicate significant differences among pomaces. Capital letters in the same column indicate significant differences among storage times. nd: not detected, and limit of detection = 1 log CFU g^{-1}. CFU: colony-forming unit. Sig.: Significance. ANOVA test significance levels: ns p>0.05; * p≤ 0.05; ** p≤ 0.01; *** p≤ 0.001.

Concerning the PPO activity, the Picual green pomaces demonstrated very little activity, and no effects were observed following the application of HHP. In contrast, the Picual mature samples exhibited higher PPO activity, with HHP treatment leading to a noticeable reduction in activity levels at both T1 and T3 (**Figure 1**).

The total phenolic (TP) content and total antioxidant activity (TAA) values (as shown in **Table 2**) were significantly higher in the green samples compared to the mature samples at all storage times. Throughout the storage period, a decline in both TP and TAA was noted in all batches, except for the green samples treated with HHP, where no changes in TP were observed during storage. Overall, the application of HHP did not have a significant effect on TP and TAA throughout the entire preservation study.

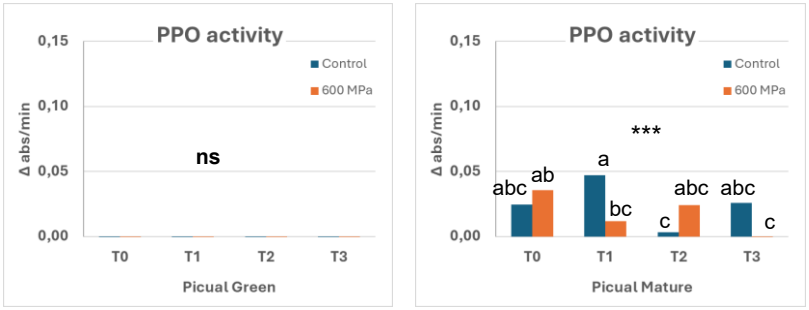

Figure 1. Polyphenol oxidase (PPO) activity of olive pomaces throughout storage for each olive type. a, b, c: different letters indicate significant differences between pomaces within a type of olive. Significance levels: ns p>0.05; *** p≤ 0.001.

Table 2. Total antioxidant activity (TAA), total phenols content (TP) of the Pical olive pomaces treated with HHP, and their evolution during storage.

		Pical green		Pical mature		
		Control	**600 MPa**	**Control**	**600 MPa**	**Sig.**
TAA	T0	76.24 ± 0.95 a A	74.87 ± 2.87 a A	68.51 ± 7.3 ab A	65.83 ± 1.69 b A	ns
	T1	58.47 ± 4.77 ab B	59.69 ± 2.14 a B	50.92 ± 2.66 bc B	47.86 ± 2.30 c B	**
	T2	54.72 ± 1.46 a B	61.13 ± 2.25 a B	43.69 ± 3.03 b BC	45.29 ± 2.94 b B	***
	T3	48.93 ± 1.15 a C	47.89 ± 1.94 a C	33.39 ± 1.57 b C	35.23 ± 1.85 b C	***
		***	***	***	***	
TP	T0	6.42 ± 0.32 a A	5.88 ± 0.36 ab	5.37 ± 0.61 ab A	4.96 ± 0.53 b A	**
	T1	5.45 ± 0.35 a B	5.68 ± 0.05 a	4.60 ± 0.06 b AB	4.39 ± 0.17 b AB	***
	T2	5.04 ± 0.26 b B	5.69 ± 0.24 a	3.87 ± 0.09 c B	3.98 ± 0.17 c B	***
	T3	5.51 ± 0.29 a B	5.46 ± 0.35 a	3.89 ± 0.21 b B	3.78 ± 0.26 b B	***
		**	ns	**	**	

TAA: total antioxidant activity (µmol Trolox equivalent g^{-1} pomace). TP: total content of phenols (mg of gallic acid equivalent g^{-1} pomace). Small letters in the same row indicate significant differences among pomaces. Capital letters in the same column indicate significant differences among storage times Sig: significance of ANOVA test. Significance levels: ns p>0.05; * p≤ 0.05; ** p≤ 0.01; *** p≤ 0.001.

The results of this work point to the possibility of using the HHP (600 MPa for 5 minutes) to control the microbiological loads of olive pomaces of Picual cultivar for their long-term preservation in refrigeration, without affecting their levels of TP and TAA, and helping to inactivate the PPO.

Acknowledgements and funding

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P7: Effect of HHP processing strategy (cycles vs continuous treatment) on colour and microbial stability of lamb burgers

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High hydrostatic pressure (HHP) is a non-thermal technology widely recognised for its ability to enhance food safety and extend the shelf life of food products while preserving their nutritional and sensory quality [1]. In meat systems, HHP is particularly effective at reducing microbial load without the need for synthetic additives, aligning with current clean-label trends. However, processing conditions such as pressure level, holding time, or treatment in cycles can significantly influence product quality, especially colour, which is a critical factor for consumer acceptance [2]. Meat products such as lamb burgers are highly perishable matrices that require effective preservation strategies due to their susceptibility to microbial growth [3]. In this context, the optimisation of HHP processing conditions is essential to balance microbial inactivation and color preservation. In some cases, two short consecutive pressure cycles are applied to enhance microbial inactivation, since the first cycle induces sublethal damage that increases microorganisms' sensitivity to the second cycle [4]. Therefore, different HHP treatments based on equivalent pressure levels, but varying time-cycle combinations may lead to distinct effects on product quality and stability over time.

Lamb burgers were prepared according to a traditional formulation using minced lamb (40% flank and 60% leg) with salt and spices, and shaped to approximately 100 g. Samples were vacuum-packaged and subjected to two HHP treatments: HHP1 (600 MPa for 1 s (2 cycles)) and HHP2 (600 MPa for 4 min), using a semi-industrial equipment (model 6000/55, Hiperbaric S.A., Burgos, Spain), with a vessel capacity of 55 L and an initial water temperature of 16 °C. Processing conditions were selected based on commercial-scale practices, where a maximum pressure of 600 MPa is commonly used, and short holding times or multiple cycles may enhance microbial inactivation. Concretely, the HHP2 treatment had an equivalent total processing time to the two consecutive cycles applied in HHP1. A total of five burgers were analysed per condition (n = 5; NO HHP, HHP1, and HHP2). After processing, samples were stored at 6 °C in darkness for 14 days and analysed at day 0 (the day after production and processing) and day 14. Non-treated burgers at day 14 were discarded due to excessive microbial growth and excluded from the experiment. The effect of HHP treatments was evaluated in terms of colour (lightness (L*), redness (a*), and yellowness (b*)) and microbiology (mesophilic counts) according to standard methods.

Table 1. Instrumental color of lamb burgers with different HHP treatments.
T0:

Storage (Days)	NO HHP	2 cycles HHP1	1 cycle HHP2	*P*-value
CIE L *				
1	46.7c ± 0.7	55.4b ± 0.8	58.1a ± 0.2	***
14	-	55.9 ± 0.5	57.0 ± 1.0	ns
P-storage		ns	*	
CIE a *				
1	8.3a ± 0.5	3.2b ± 0.2	2.7b ± 0.3	***
14	-	3.1 ± 0.4	2.3 ± 0.3	**
P-storage		ns	*	
CIE b *				
1	13.2a ± 0.4	11.9b ± 0.5	11.8b ± 0.5	***
14	-	13.1 ± 0.2	12.7 ± 0.2	*
P-storage		***	**	

initial T1: 14 days. HHP1: 600 MPa 1 s (2 cycles). HHP2: 600 MPa 4 min. Values are expressed as mean ± standard deviation. Different letters within each parameter indicate significant differences between treatments according to Tukey's test ($p \leq 0.05$). * Significance at $p \leq 0.05$. ** Significance at $p \leq 0.01$; *** Significance at $p \leq 0.001$; ns non-significant differences.

HHP treatments significantly affected the instrumental colour of lamb burgers (**Table 1**). At day 1, both treatments increased L* compared to untreated samples, with higher values observed for HHP2 (1-cycle) followed by HHP1 (2-cycles), and lower values in NO HHP. CIE a* and b* were significantly reduced after both HHP

treatments, with no significant differences between them at this time point. After 14 days of refrigerated storage, both treatments maintained similar L* values. However, significant differences in a* and b* were observed, with HHP1 (2-cycles) showing higher values than HHP2 (1-cycle). Storage effects depended on the treatment. In HHP1 (2-cycles) burgers, L* and a* remained stable, while b* showed higher values. In contrast, HHP2 (1-cycle) samples, storage resulted in a decrease in L* and a*, together with an increase in b*, indicating a progressive loss of redness and a less vivid appearance. HHP treatment clearly influenced the visual appearance of lamb burgers. While both treatments modified colour, HHP2 (1-cycle) produced burgers with a paler and less characteristic appearance. In contrast, HHP1 (2-cycles) better maintained the colour of lamb burgers, resulting in a more visually appealing product over time. Regarding mesophilic counts of lamb burgers with different HHP treatments (**Table 2**), both HHP treatments effectively reduced mesophilic counts compared to NO HHP at day 0. After 14 days of refrigerated storage, similar microbial levels were observed in both treatments, indicating no differences between HHP1 (2-cycles) and HHP2 (1-cycle). Storage did not affect mesophilic counts in HHP1 samples, while a slight increase was observed in HHP2 (1-cycle).

Table 2. Mesophilic counts (log colony forming units CFU g^{-1}) of lamb burgers with different HHP treatments.

Mesophilic counts (log CFU g^{-1})				
Storage (Days)	**NO HHP**	**2 cycles HHP1**	**1 cycle HHP2**	***P-value***
1	7.4a ± 0.1	4.1b ± 0.3	3.7b ± 0.3	***
14	-	4.7 ± 0.8	4.7 ± 0.5	ns
p-storage	-	ns	**	

T0: initial T1: 14 days. HHP1: 600 MPa 1 s (2 cycles). HHP2: 600 MPa 4 min. Values are expressed as mean ± standard deviation. Different letters within each parameter indicate significant differences between treatments according to Tukey's test ($p \leq 0.05$). ** Significance at $p \leq 0.01$; *** Significance at $p \leq 0.001$; ns non-significant differences.

The results demonstrate that HHP treatment has a clear impact on the visual quality of lamb burgers. While both treatments modified colour, the multi-cycle treatment better preserved the typical fresh appearance of the product, whereas the single longer treatment led to a more pronounced loss of red colour and a less visually appealing product during storage. In terms of microbiological quality, both HHP treatments showed similar effectiveness, maintaining comparable levels of microbial control throughout storage and preventing microbial growth over time.

These findings highlight that HHP treatments based on short cycles can achieve the same microbiological control as continuous HHP treatments, while minimizing color deterioration. This approach represents a more suitable strategy for extending shelf life without compromising product appearance.

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P8: Effect of high hydrostatic pressure on microbial inactivation and functional compounds in red pepper by-products

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Red pepper (*Capsicum annuum* L.) is an important crop in Spain, particularly in the Extremadura region. During processing and commercialization, a significant proportion of peppers is discarded as by-products due to not meeting required size, shape, or visual quality standards, despite being fully edible. These by-products are currently underutilised, even though they are rich in bioactive compounds such as carotenoids and phenolics [1]. However, their high moisture content and pH make them highly perishable, promoting rapid microbial spoilage and limiting their direct use as food ingredients. In parallel, current strategies for agri-food by-product valorisation are largely based on fractionation and extraction processes, which may involve additional costs, solvent use, and the generation of secondary waste. In contrast, the use of Hydrostatic high pressure (HHP) as a strategy for direct stabilisation of by-products, preserving the whole matrix without extraction processes, remains largely unexplored. HHP is a non-thermal technology that allows the preservation of food products [2]. This technology reduces microbiological counts while maintaining and even increases levels of bioactive compounds [3]. This approach offers a more sustainable and integrative alternative, enabling direct utilisation while maintaining structural and functional properties [4]. Therefore, the aim of this study was to evaluate the application of HHP (600 MPa, 5 min) as a valorisation strategy for red pepper by-products, focusing on its effect on microbiological quality, as well as on key functional compounds, in order to assess their potential use as natural ingredients in food systems.

Red pepper (*Capsicum annuum* L.) by-products were supplied by a local company and processed into a homogeneous purée. The purée was vacuum packaged in plastic bags and subjected to HHP treatment at 600 MPa for 5 min using a semi-industrial system (model 6000/55, Hiperbaric S.A., Burgos, Spain), with a vessel capacity of 55 L and an initial water temperature of 16 °C. First, the physicochemical composition of the purée was analyzed using descriptive statistics. The effect of HHP on microbiological (ISO standards), total phenolic compounds (TPC) (Folin-Ciocalteu method), total carotenoid content (TCC) (UV–Vis spectrophotometry), and antioxidant activity (AA) (ABTS assay) was evaluated by one-way ANOVA and Tukey-b test $p \leq 0.05$). For quantitative analysis of microbiology, counts below detection limits (<1 log CFU g^{-1}) were assigned values of 0.99.

Table 1. Physicochemical composition of the red pepper by-product.

pH	5.2 ± 0.0
Protein (g 100g^{-1})	1.1 ± 0.0
Moisture (g 100g^{-1})	91.4 ± 0.6
Fiber (g 100g^{-1})	1.9 ± 0.2

The values are presented as the mean ± standard deviation

The red pepper by-products presented a high moisture content (91.4 g 100g^{-1}) and acidic pH (5.2), as seen in **Table 1**, which are relevant factors for their stability and potential application. HHP treatment at 600 MPa for 5 min resulted in a significant reduction in microbial load (**Table 2**), with mesophilic and psychrophilic counts, as well as moulds and yeasts, decreasing from initial levels of 6.5, 6.2, and 4.0 log CFU g^{-1}, respectively, to below the detection limit (<1 log CFU g^{-1}).

Table 2. Microbiological counts (log CFU g^{-1}) changes in high-pressure treated red pepper by-product purée

	Untreated	600 MPa 5 min	*P-treatment*
Mesophilic	6.5 ± 0.0	<1 ± 0.0	***
Psychrophilic	6.2 ± 0.1	<1 ± 0.0	***
Moulds and yeasts	4.0 ± 0.0	<1 ± 0.0	***

The values are presented as the mean ± standard deviation. <1: non-detected, below detection limit (<1 log CFU g^{-1}). Significant differences between treatments were determined by one-way ANOVA and Tukey-b post-hoc test. *** $p \leq 0.001$.

Regarding functional properties (**Figure 1**), TCC was not significantly affected by HHP treatment (146.5 vs 137.9 µg BCE g^{-1}, $p > 0.05$). In contrast, TPC showed a significant decrease after processing (1384.6 *vs.* 1098.5 µg GAE g^{-1}, $p < 0.01$). However, AA significantly increased (2.6 vs 2.8 mg TE g^{-1}, $p < 0.05$), suggesting possible changes in the extractability or interaction of antioxidant compounds within the matrix.

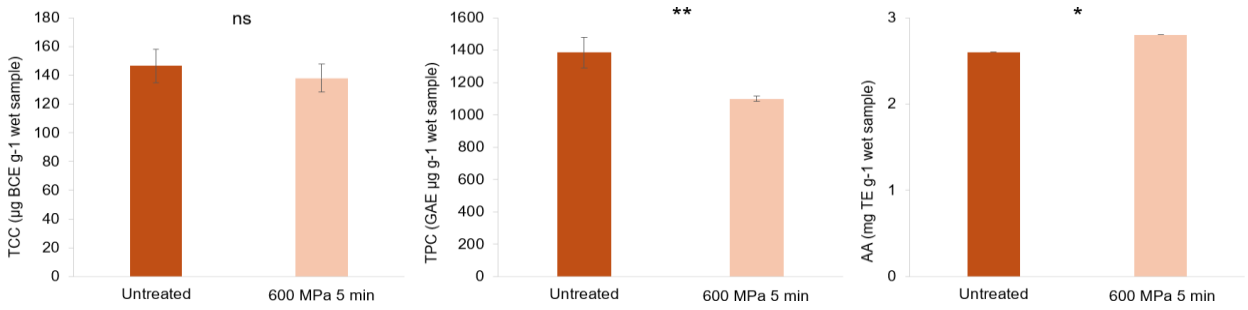

Figure 1. Changes in TCC (µg β-carotene equivalents (BCE) g^{-1} wet sample), TPC (µg gallic acid equivalents (GAE) g^{-1} wet sample), and AA (mg Trolox equivalents (TE) g^{-1} wet sample) in high-pressure treated red pepper by-product purée. The values are presented as the mean ± standard deviation. Significant differences between treatments were determined by one-way ANOVA and Tukey-b post-hoc test. ns: non-significant differences. *$p \leq 0.05$. **$p \leq 0.01$.

The results demonstrate that HHP (600 MPa 5 min) is an effective strategy for the direct stabilisation of red pepper by-products, achieving substantial microbial inactivation to levels below detection limits while preserving key functional properties of the matrix. Despite a reduction in TPC, the stability of TCC and the significant increase in AA suggest that HHP induces structural modifications that enhance the accessibility of bioactive compounds. Importantly, this study provides novel evidence supporting the application of HHP as a sustainable alternative to conventional valorisation strategies, enabling the stabilisation of agri-food by-products without the need for extraction or fractionation processes. This approach preserves the integrity of the whole matrix and facilitates its direct incorporation as a natural ingredient in food systems, contributing to the development of clean-label and circular economy solutions.

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P9: Valorisation of chilli peppers by-products using high-pressure as an extraction technology

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The fruits of the *Capsicum* species, commonly called piquant peppers or chili peppers, are characterized by their pungency due to the presence of capsaicinoids (CAP) and dihydrocapsaicin (DHC). The industrial process of spicy sauces results in a spicy byproduct (pepper pomace) that requires additional treatment, as its disposal is harmful to the local ecosystems and presents composting challenges.

Currently, conventional CAP and DHC extraction processes are based on the application of high temperatures for long periods to increase extraction yield. However, many of these compounds degrade rapidly, and the use of temperature represents an economic and environmental disadvantage. Non-thermal processing technologies, such as high-pressure processing (HPE), have demonstrated enormous potential for their extraction, as it is carried out at room temperature (RT), without degrading the heat-sensitive compounds.

Thus, the objective of this work was to apply new extraction strategies for compounds such as CAP and DHC using HPE to reduce the pungency (measured in Scoville Heat Units, SHU) of the spicy pepper pomace byproduct.

HPE assays were performed sequentially, starting with a water pretreatment (400 MPa/6 min/RT), followed by a treatment with 96% ethanol (600 MPa/6 min/RT), in order to evaluate whether this combination equals or surpasses the SHU level and the CAP and DHC content, compared to conventional extraction (80 ºC, 5 h, 96 % ethanol). The HPE-based extraction process resulted in an oleoresin with mass yields similar to the conventional temperature-based process, but with higher SHU values, obtaining a less pungent pepper pomace byproduct.

Therefore, the use of HPE resulted in a process that promotes the circular economy, adding value to byproducts that would otherwise be wasted.

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P10: Hyperbaric storage at room temperature of raw poultry meat as a novel preservation methodology compared to refrigeration

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Global poultry meat consumption is expected to continue the upward trend of the last decade, with a projected growth of 21 % by 2034, accounting for 45 % of the protein consumed from all meat sources. Poultry meat consumption rising is due to its low cost, rich nutritional profile (protein-to-fat ratio) and reduced greenhouse gas emissions, making it more appealing to health- and sustainability-conscious consumers[1]. Alongside food safety, poultry meat quality encompasses various characteristics, with colour, texture, juiciness, wateriness, odour, and flavour being some of the most important parameters for consumers before and after purchasing a meat product[2,3].

Conventional preservation methodologies such as refrigeration (RF) have been shown to be not fully effective in maintaining the microbiological safety of poultry meat and to prevent quality changes during storage. Although, current pasteurisation methodologies, like high-pressure processing (HPP), effectively inactivate the microbial load but considerably alter some physicochemical properties such as colour (giving a cooked-like appearance) and texture, making raw meat less appealing to consumers[4]. Hyperbaric storage (HS) has recently been shown to be a food preservation method that uses low/moderate hydrostatic pressures to hurdle microbial growth, even achieving pasteurisation-like levels, but with less impact on quality parameters when compared to HPP of raw meat and a longer shelf-life compared to refrigeration.

This work evaluated the impact of HS at room temperature (100-125 MPa, up to 28 days, 20-25 ºC), on the quality and safety of vacuum-packed raw chicken breast meat compared to RF and HPP (400 MPa, 5 min). Therefore, the main groups of endogenous microorganisms were evaluated (total aerobic mesophilic, Enterobacteriaceae and lactic acid bacteria) along with quality parameters such as texture, protein solubility (total, sarcoplasmic and myofibrillar proteins), and protein carbonyl.

The results showed that both HS/RT (100 and 125 MPa) conditions were not only effective in avoiding microbial growth but also resulted in spoilage microorganisms' inactivation during storage as was HPP, while under RF microbial growth exceeded consumption limits after 7 days (> 6.5 logs for mesophiles). Texture analysis revealed a general tenderization effect along storage for HS samples (approximately 35% less hardness on day 28), contrasting with the HPP samples' increased hardness (around 65%), while refrigerated samples showed constant values. Regarding to the solubility of total, sarcoplasmic and, myofibrillar proteins, values remained similar to the initial point (150 mg/g meat for total proteins) in the refrigerated samples, in opposition to a significant decrease in solubility for HPP-treated samples (32 mg/g meat for total proteins). HS/RT samples generally showed a decreasing tendency that become more evident after 3 days of storage, especially for 125 MPa (on day 28, the total protein obtained was 72 mg/g meat). Finally, the quantification of protein carbonyls revealed that both preservation methodologies promoted protein oxidation during storage (bellow 2.80 nmol/mg protein at 28th day).

The results of this work point to the possibility of using HS/RT to control the development of spoilage microorganisms, and even inactivate them at RT, in opposition to refrigeration that only slowed down microbial development, and preventing undesirable quality changes during storage, compared to HPP. Therefore, HS/RT can be considered to preserve raw chicken meat for up to 28 days with higher microbial safety and longer shelf-life, making it more appealing for consumers, being in addition *quasi*-energetically costless and so, potentially with lower greenhouse gas emissions and more sustainable.

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P11: Optimization of the extraction process of chilli peppers by-products using high-pressure

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Hot or chili peppers, fruits of the *Capsicum* species, have a high level of pungency due to the presence of compounds such as capsaicinoids (CAP) and dihydrocapsaicin (DHC). These peppers are widely used by the food industry to produce spicy sauces, resulting in a spicy byproduct (pepper pomace) that requires additional treatment since it is harmful to the environment and presents composting challenges.

Conventional extraction processes used to recover CAP and DHC rely on high temperatures that facilitate the removal and dissolution of impurities, resulting in a spicy oleoresin with low purity, making CAP/DHC quantification difficult. Non-thermal processing technologies, such as high-pressure extraction, are recognized for their use in plant matrices to disrupt vegetal cells, promoting the dissolution of cellular contents and impurities, thus emerging as an interesting new possibility for the extraction of CAPs/DHCs.

Therefore, the objective of this study was to apply new extraction strategies using high-pressure extraction (HPE) as a pretreatment step for the pepper pomace byproduct, aiming the removal of impurities and salts, so that, in the subsequent conventional extraction process, an oleoresin of higher purity is obtained, maximizing the reduction of pungency (measured in Scoville Heat Units, SHU) of the pepper pomace byproduct.

The extraction assays were conducted sequentially, beginning with HPE at 400 MPa for 6 minutes at room temperature, using water as solvent, followed by temperature-based extraction (80 °C, 1 hour, 96 % ethanol).

The application of HPE as an initial extraction step resulted in an oleoresin with a higher degree of purity (%), maintained mass yields (%), and higher CAP/DHD content, and thus an oleoresin with higher SHU values. Notably, this yielded a less pungent chilli pepper byproduct compared to the isolated application of conventional temperature-based extraction.

Thus, the use of HPE as a pretreatment allowed the optimization of the CAP/DHD extraction process, resulting in a purer and spicier oleoresin, as well as promoting a circular economy through the valorisation of chilli pepper byproducts that would otherwise be wasted.

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P12: Preservation of egg white at room temperature by hyperbaric storage: impact on volatile profile and sensory attributes

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Nonthermal pressure-based preservation technologies, specifically room temperature hyperbaric storage (HS/RT) and moderate pressure pasteurization (MPP), represent a possible new paradigm in food preservation. Operating at mild pressures (up to 200 MPa), HS/RT and MPP offer a critical advantage by effectively inhibiting and inactivating pathogens without compromising in great extent quality. These methodologies have particularly interest for highly perishable matrices like egg products, where conventional thermal processing (TP) not only degrades important functionalities but along with subsequent refrigeration (RF, 4ºC) imposes significant economic and environmental issues[1].

Hen egg white (EW) is important food industry ingredient due to its rich protein content and its ability to impart desirable functional properties, although it represents a significant salmonellosis risk necessitating preservation methodologies[2]. Recent HS/RT and MPP studies showed that, using pressures up to 200 MPa, showed to effectively inhibit/inactivate various *Salmonella* spp., and gram-positive bacteria such *Listeria monocytogenes* and *Staphylococcus aureus*.

As a complex food ingredient, EW is highly susceptible to flavour alterations during processing and storage. These preservation methods can significantly modify its volatile organic compound (VOC) profile, ultimately impacting sensory attributes and consumer acceptance. Evaluating these changes is essential to understand the chemical modifications and thereby optimizing processing parameters to preserve the EW flavour characteristics[3].

Therefore, VOC profile was evaluated on EW stored under HS/RT (50 MPa up to 60 days) and pasteurized by MPP (175 MPa/1 day), with the respective non-pasteurized refrigerated (RF, 4ºC) and TP controls. Changes in the volatile profile were assessed by headspace solid-phase microextraction (HS-SPME) coupled to gas chromatography-mass spectrometry (GC-MS). Also, sensory attributes were evaluated on raw EW, with semi-trained panel performing a quantitively descriptive analysis (QDA).

The application of nonthermal preservation methodologies modified the volatile organic compound (VOC) profile of EW. HS/RT induced the most significant increase ($p<0.05$), with total VOC content after 35 days measuring 4-times higher than RF samples, an elevation that continued until the 60 days of storage. This rise was mainly due to the increase in alcohols (94 % increase after 35 days, comparing to RF), aldehydes (hexanal and nonanal) and toluene. In addition, for both HS/RT and MPP, ester content decreased whereas nitrogenous compounds increased compared to EW prior storage and RF. Concerning MPP, volatile profile remained similar to TP, with PCA clustering both conditions together.

Sensory evaluation demonstrated that despite these volatile changes, odour and heterogeneity, remained similar between conditions, including the global acceptability, with viscosity and colour being the primary differentiators attribute detected by panellists.

Thus, HS/RT maintain or enhance key aroma attributes, although not perceived by semi-trained panellists. These results highlight HS/RT and MPP as a low-energy, climate-friendly preservation methodologies with clear potential for its application in EW.

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P13: Food Safety Assessment of Almond Ice Cream Formulated with Liquid Fresh Whey: Comparison between High Pressure Processing and Thermal Pasteurization

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The valorization of liquid fresh whey in food formulations aligns with circular bioeconomy strategies that aim to convert nutrient-rich side streams into value-added products [1]. In this context, almond ice cream formulated with whey represents a promising opportunity to combine product innovation with improved resource efficiency. However, integrating dairy co-products into the food value chain while ensuring microbiological safety remains challenging, particularly when minimally processed ingredients are used.

High pressure processing (HPP) is a nonthermal technology typically operating at industrial conditions of 400–600 MPa for short treatment times (3–5 minutes). It can ensure microbial safety by inactivating vegetative microorganisms and reducing enzymatic activity, while better preserving sensory and nutritional quality than conventional thermal pasteurization [2].

This study aimed to evaluate the valorization of liquid fresh whey through its microbiological stabilization and subsequent incorporation into an almond ice cream formulation. In addition, the work sought to perform a pilot-scale validation of the formulation and preservation strategies, comparing conventional thermal pasteurization and HPP in terms of microbiological safety at day 7 post-production.

Liquid fresh whey was stabilized either by thermal treatment (70 °C, 20 minutes) or by HPP (600 MPa, 5 minutes, 18 °C), and its microbiological stability under refrigerated storage was confirmed. These treatments aimed to inactivate enzymes and reduce the initial microbial load, preventing fermentation or degradation prior to its incorporation into the ice cream formulation.

The almond ice cream formulation included 50% pre-stabilized whey. The ice cream mix was subsequently processed using two different preservation technologies: conventional thermal pasteurization (90 °C, 13 minutes) and nonthermal HPP (500 MPa, 4 minutes, 18 °C), in accordance with food safety requirements for compound dairy-based products. In both cases, the impact of the processing technology on microbiological safety was evaluated at day 7.

The results obtained for the whey fraction confirmed that HPP (600 MPa, 5 min) allowed its microbiological stabilization under the conditions used, supporting its incorporation at 50% of the formulation as a safe ingredient. After 7 days of both thermal (90 °C, 13 minutes) and HPP (500 MPa, 4 minutes) pasteurization, the almond ice cream showed low total viable counts at 30 °C (in the order of 10^2–10^3 CFU/g), yeasts and moulds counts below the detection limit, as well as the absence (in 25 g) of *Salmonella* spp. and *Listeria monocytogenes*, with Enterobacteriaceae and B*acillus cereus* counts below the guideline values for ice cream and frozen desserts, indicating that both preservation strategies ensured satisfactory microbiological quality of the product [3].

Taken together, these results support the use of HPP, in combination with HPP-stabilized whey, as a viable approach for producing almond ice cream with microbiological safety and added theoretical nutritional interest, contributing also to the valorization of a co-product like whey, reintegrating it as a food ingredient. In addition, HPP allowed to reduce the pasteurization times compared to thermal pasteurization, which can be an interesting alternative from an industrial standpoint, as it achieves similar levels of microbial inactivation with shorter processing times.

Rheological and texture analyses remain part of the ongoing work. They will be used to assess how thermal and high-pressure treatments influence the flow behaviour of the ice cream mix as well as the textural properties and structure of the almond ice cream, and the results will be presented at the conference.

Acknowledgements and funding

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P14: Antioxidant and antimicrobial activities of seaweed extracts obtained by high-pressure assisted extraction

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In recent years, the demand for functional products, particularly natural ingredients, has significantly increased, with consumers showing a clear preference for these over synthetic alternatives. This trend is growing not only in the food industry but also across other sectors. In this context, seaweed has emerged as valuable food resources due to their nutritional and chemical composition [1]. Macroalgae are rich in mineral elements (7–36%), lipids (1–5%), polysaccharides (15–76%), and proteins (5–47%), with these values referring to the fresh weight of the algae [2, 3]. Particularly, polysaccharides extracted from macroalgae, such as agar, alginate, and carrageenan, have garnered significant attention for their applications as thickening, stabilizing, and emulsifying agents in the food industry [4, 5]. Although they have a low lipid content, macroalgae exhibit a high concentration of polyunsaturated fatty acids (PUFAs) and other lipid compounds that provide health benefits [6, 7]. Moreover, they contain significant amounts of micronutrients such as vitamins and antioxidant secondary metabolites, including polyphenols and pigments [8]. The wide range of bioactive molecules identified in macroalgae has driven numerous studies, which have demonstrated their biological properties, such as anti-inflammatory, antioxidant, antimicrobial, antidiabetic, anticancer, neuroprotective, and photoprotective activities, among others [9, 10]. Given the growing interest in identifying compounds derived from macroalgae, it is crucial to develop efficient, sustainable, and economically viable processes for their extraction [11]. While some of these compounds can be chemically synthesized, extracting bioactive compounds using green extraction technologies represents a more cost-effective and environmentally friendly alternative, which also avoids the use of hazardous chemical substances. The extraction of bioactive compounds is an essential step in recovering these molecules from plant matrices. Presently, high hydrostatic pressure is increasingly being explored as a nonthermal extraction method (called high pressure-assisted extraction, HPE) for bioactive compounds from natural materials. The aim of an efficient extraction procedure is to provide the maxi-mum yield of substances with the highest functional properties, enhance the bioactivity of the extracts, increase pollution prevention, and shorten extraction time [12]. HPE is considered one of the most environmentally friendly technologies. It can recover target compounds is a shorter time, and the process can be carried out at room temperature or below thereby avoiding the thermal degradation of thermosensitive molecules.

This study aimed to investigate the antioxidant and antimicrobial properties of extracts obtained from five macroalgae species using HPE with a view of their incorporation in coatings to extend the shelf-life of seafood products. Two extraction cycles were carried out at 500 MPa for 3 and 10 minutes. The macroalgae species used were *Bifurcaria bifurcata*, *Codium tomentosum*, *Fucus spiralis*, *Porphyra dioica* and *Palmaria palmata* and the extracts were evaluated for polyphenol content, antioxidant activity (DPPH, FRAP and ABTS assays) and antimicrobial activity against *Escherichia coli*, *Listeria innocua*, *Staphylococcus aureus*, *Bacillus subtilis*, *Pseudomonas putida* and *Shewanella putrefaciens*. HPE demonstrated high efficiency with yields of approximately 50% using distilled water as a solvent, making the process safe, sustainable and easily scalable. The results obtained demonstrate that the most promising extracts in terms of antioxidant and antimicrobial activities are in extracts of F. spiralis with 10 and 3 min of extraction. The 10-minute extract exhibited the highest total phenolic content (80.93 ± 15.09 mg GAE/g extract) and the strongest antimicrobial effect against *S. putrefaciens* (13.69 ± 4.42% growth), whereas the 3-minute extract showed the best antioxidant performance, with DPPH activity of 55.49 ± 11.20% and FRAP reducing capacity of 59.7 ± 13.61 µmol AAE/g. Therefore, a combination of these two extracts could be a good option to be included in a coating formulation with the aim of increasing the shelf life of fresh fish products.

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Cells and Organisms and Biotechnology Applications under High Pressure

O11: Room temperature hyperbaric storage of human adipose derived stem cells as a potential alternative to cryopreservation

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Stem cells are fundamental for cell-based therapies and regenerative medicine due to the self-renewal and differentiation capacity[1]. Effective preservation strategies are critical to maintain their therapeutic potential. While cryopreservation remains the gold standard for extending cell lifetime and storage, it presents limitations such as the need for cryoprotective agents (e.g. DMSO), risk of cryoinjuries, and high energy consumption[2–4]. Hyperbaric storage (HS) is an emerging preservation technique with potential for preservation of foods and biological products under pressure[5]. This method has been mainly studied in food products, with results demonstrating the potential to extend shelf life and maintain quality parameters in several food matrices[6–8]. When applied at room temperature (RT), HS can be considered a quasi-energetically costless procedure, as no additional energy is needed during storage after the initial pressurization, leading to significant energy savings[9]. This study evaluates the feasibility of HS at RT as a novel, cryoprotectant-free, and low-energy alternative for preserving stem cells using human adipose stem cells as a relevant study model (**Figure 1**).

Cells were stored under pressure (20, 30, and 40 MPa) at RT, up to 21 days, and compared to cells preserved at −80 °C. To confirm the potential of HS as a cell preservation method, in vitro characterization assays were done, namely, cell viability, metabolic activity, DNA quantification and phenotype determination.

After 3 days of storage, cells kept at 30 MPa/RT presented the best results with similar viability, and metabolic activity compared to cryopreserved controls while phenotypic markers were maintained. Moreover, cells stored under HS (despite the pressure used during storage) exhibited a shorter adaptation period post-storage, indicating potential advantages for rapid downstream applications. Results after 7 days of storage showed a reduction in metabolic activity and viability, however phenotypic markers were maintained and cells kept normal morphology for adherent hASCs. Storage for longer periods (14 and 21 days) resulted in reduced viability and adherence, although stemness markers remained similar throughout storage timeframe.

These findings suggest that short-to-mid-term storage under HS/RT (30 MPa/RT) can preserve stem cells function effectively with low energy costs and without the need for additives such as cryoprotective agents. This initial evaluation supports the feasibility of HS/RT as a possible alternative to conventional cryopreservation, particularly for applications involving transport or short-term storage and paves the way for additional research in this field.

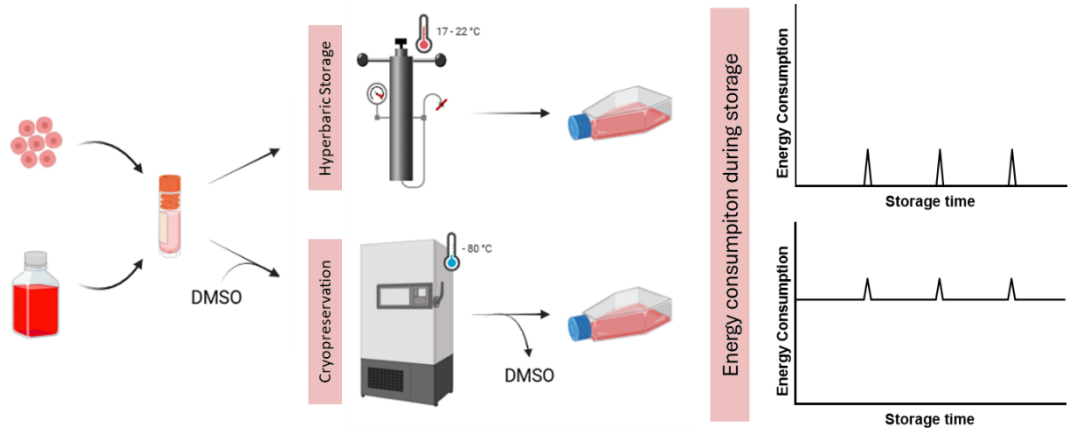

Figure 2. Graphical overview of human adipose-derived stem cells (hASCs) storage using two approaches: additive free hyperbaric storage at room temperature (HS/RT) and conventional cryopreservation (- 80 ºC) with DMSO. The comparison includes a qualitative energy consumption profile, illustrating the overall energy demand of each method over time, with the energy peaks spotlighted for each method corresponding to three hypothesized depressurization/opening and pressurization/closing events during the storage period.

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O12: Development of high pressure transformation of the haloalkaliphilic archaeon *Natronomonas pharaonic*

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The development of green chemistry to meet future needs in industrial processes relies in part on the use of extremozymes. Long produced in *E. coli* or other mesophilic organisms, several of these extremozymes have shown little or no activity, stressing the need for appropriate extremophilic protein production organisms (cellular chassis). Haloalkaliphilic Archaea, thriving in hypersaline (20-30% NaCl) and hyperalkaline (pH > 9) environments, constitute valuable models for fundamental research and promising resources for biotechnological applications for use in extreme saline and alkaline processes. One of the most promising haloalkaliphilic chassis is *Natronomonas pharaonis*. However, its use remains limited by the lack of efficient genetic tools and transformation protocols, which limits both func-tional studies and industrial development.

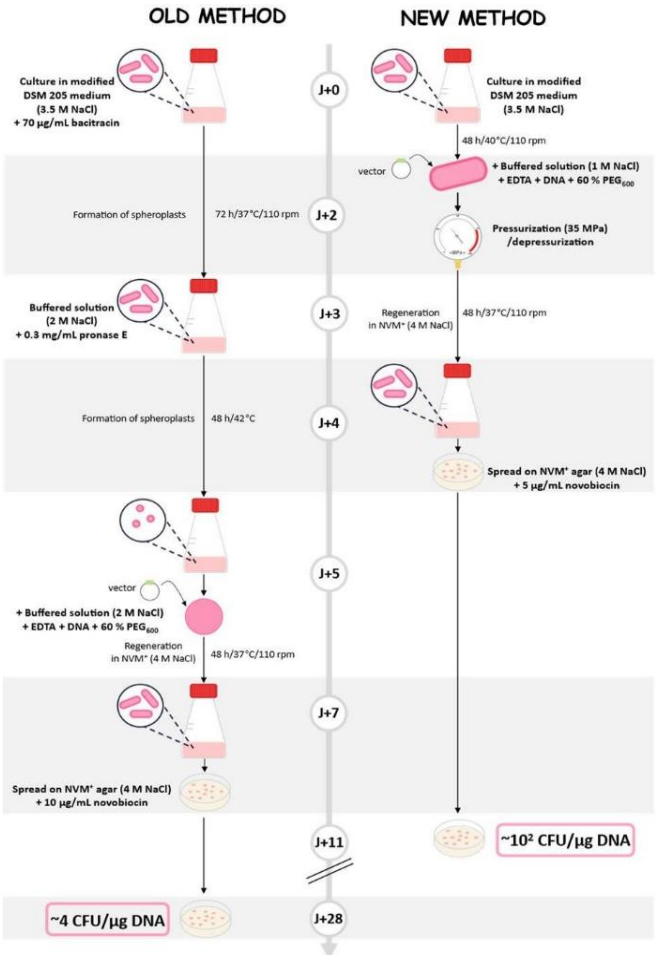

Figure 1. PEG-mediated spheroplast transformation (left) vs. HHP-based (right) protocols.

To date, only a few vectors have been reported [1] and a single transformation procedure, whose efficiency is very limited (ca. 4 CFU/µg of DNA). In comparison, trans-formation of *E. coli* reaches up to 10^9 CFU/µg of DNA. The current transformation method relies on Polyethylene Glycol (PEG)-medi-ated spheroplast transformation (**Figure 1, left**) [1]. Although this approach is simple, re-producible, and robust in halophilic Archaea, it is still very time-consuming, requiring 28 days from start to transformant.

Here, we present a novel transfor-mation method for *Nmn. pharaonis* based on the application of high hydrostatic pressure (HHP) followed by rapid depressurization (**Figure 1**) [2]. Rapid HHP release has recently emerged as a promising tool for DNA deliv-ery in difficult-to-modify primary cells, includ-ing embryonic stem cells[3].

Here, we show that applying 35 MPa enables efficient transformation of *Nmn. pharaonis*. HHP and release were applied as an extra step in the protocol after the for-mation of spheroplasts, but yielded no transformants, as well as no survival of the spheroplast to the HHP treatment.

The same approach was tested on intact cells, omitting the formation of spheroplasts. Transformation efficiency was increased by nearly two orders of magnitude compared to the classic PEG-mediated method (400 vs 4.1 CFU/µg DNA) [2]. In further optimizations, we tested the impact of a temporary decrease in the NaCl concentration from the optimal 4 M for growth to up to 0.5 M during DNA exposure. Low NaCl concentrations create an osmotic shock in the cells and can help to fragilize the cell membranes. However, it also drastically reduces cell viability. Remarkably, the reduction from the optimal 4 M for growth to 1 M did not impair cell survival, nor transformation efficiency (**Figure 2**). However, it allowed transformants to appear sev-eral weeks earlier than at lower or higher salinities. First transformants can be subcultured starting at day 7, reducing the time required for transformation from 28 days following the classic PEG-mediated spheroplast transformation, to 11 days using HHP[2].

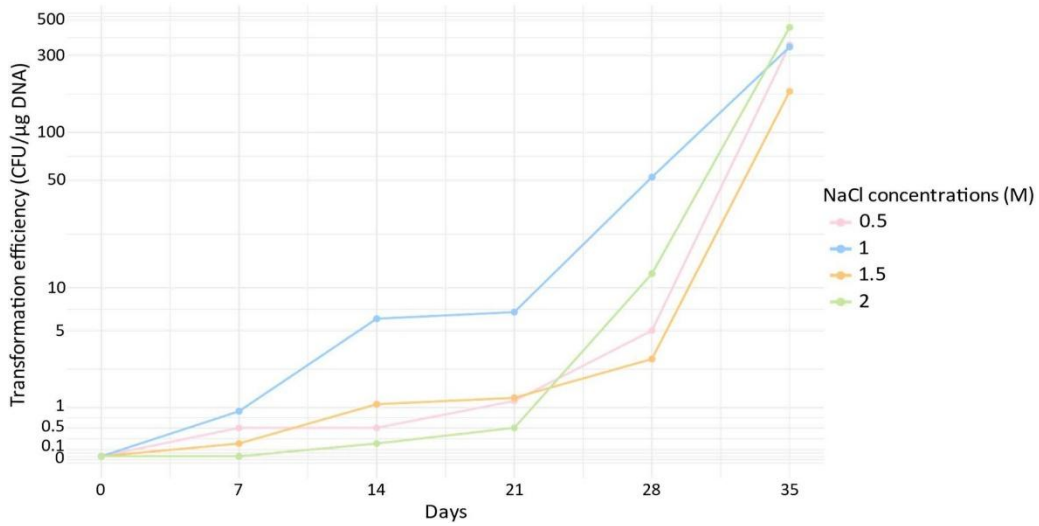

Figure 2. Effect of NaCl concentration on transformation efficiency over time. Transformation assays were performed 35 MPa in triplicates.

We attribute the improvement of transformation efficiency to pressure-induced membrane reorganization, promoting transient pore formation, consistent with previously reported effects of HHP on archaeal and bacterial membranes. Overall, this method is likely to be transposable to other recalcitrant Archaea, opening new perspectives for both fundamental research and industrial applications.

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O13: Membrane adaptation to high pressure involves bulk lipid transfer at ER–PM contact sites

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Biological membranes must maintain their physical properties to support protein function and cellular viability under environmental stress. Hydrostatic pressure provides a well-defined physical perturbation that directly constrains lipid packing and increases membrane order, offering a useful framework to examine membrane adaptation mechanisms. However, how high pressure influences lipid synthesis and the complex membrane trafficking systems operating in living cells remains unclear.

Here, we used the budding yeast *Saccharomyces cerevisiae* as a genetically tractable eukaryotic model and carried out a genome-wide screen using the yeast deletion collection to identify genes required for growth under nonlethal high-pressure conditions (~25 MPa)[1]. Among the mutants identified, we focused on *CSF1*, a gene whose function remains incompletely understood. *CSF1* was originally identified based on a cold-sensitive fermentation phenotype, but its deletion causes a severe growth defect under high pressure[1]. Recent studies suggest that Csf1 is a tunnel-like lipid transfer protein[2]. Its human homolog, BLTP1, is associated with Alkuraya–Kučinskas syndrome, a severe genetic disorder.

To examine the role of Csf1 in membrane adaptation, we compared lipid composition and membrane physical properties between wild-type and *csf1*Δ cells. Quantitative lipidomics combined with fluorescence anisotropy measurements showed that Csf1 is required for coordinated lipid remodeling under pressure[3]. In its absence, phospholipid unsaturation is broadly reduced, leading to increased rigidity of the endoplasmic reticulum (ER) membrane and destabilization of plasma membrane (PM) proteins, particularly amino acid permeases[3]. These results indicate that membrane environments compatible with protein function cannot be maintained without Csf1 under high pressure.

Consistent with our previous studies[4,5], deletion of the ER membrane protein Ehg1 results in closely related defects under pressure, supporting a functional link between Csf1 and Ehg1 in maintaining membrane integrity. These observations suggest that Csf1 operates within a membrane-associated system that sustains lipid dynamics under stress, likely at ER–PM contact sites.

Together, these findings link bulk lipid transfer to the maintenance of membrane physical properties under pressure. More broadly, they provide a framework for understanding how eukaryotic cells coordinate lipid transport and membrane biophysics to remain functional under extreme physical conditions, with implications for deep-sea biology, membrane protein stability, and stress adaptation.

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O14: Expanding the Pressure Frontier in Grüneisen Parameter Measurement

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The Grüneisen parameter (γ) is crucial for determining many thermal properties, including the anharmonic effect, thermostatistics, and equation of state (EOS) of materials. This relationship is critical in both life sciences and material sciences. For example, Reid et al. demonstrated for a small adjustment in average volume, due to a change in state of a protein or other macromolecule at constant temperature, the change in vibrational entropy is related to the mode Grüneisen parameters, which relate shifts in frequency to a small volume change[1]. Similarly, in high-pressure physics, the Grüneisen parameter governs the anharmonicity and the thermal pressure of solids, yet its direct experimental determination remains challenging due to the difficulty of achieving isentropic compression. In this work, we overcome this obstacle by developing a new technique capable of generating quasi-isentropic adiabatic compressions up to tens of GPa within an extremely short time (~0.5 ms) using a dynamic diamond anvil cell (dDAC). We applied this method to sodium chloride and successfully determined its γ over a wide pressure range. The calculated Hugoniot curve of the B1 phase of NaCl, derived from our measured γ values, reaches up to 20 GPa and 960 K and is in excellent agreement with previous shock-compression experiments (e.g., Fritz et al.[2]).

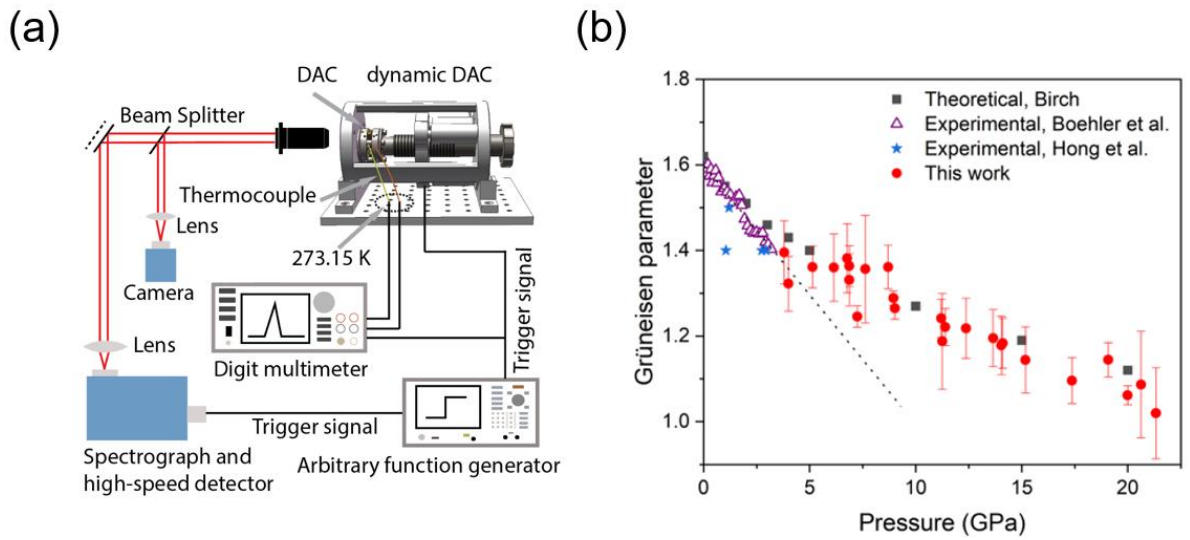

Figure 1. (a) Schematic illustration of the programmable automatic high pressure jump and temperature detection apparatus[3]. (b) Grüneisen parameter vs. pressure for sodium chloride from our data and previously published data[4-6]. We measured the Grüneisen parameters in a wide pressure range. The dashed line is the extrapolation from the previous experimental data.

Our findings confirm that this new method reliably measures the Grüneisen parameter of solids under extreme conditions. Taken together, works on proteins and on sodium chloride underscores the general and powerful role of the Grüneisen parameter in bridging microscopic vibration to macroscopic thermodynamic observables across diverse fields, from molecular biology to condensed matter physics.

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O15: High pressure treatment of cellular assemblies towards stable therapeutic products

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Mesenchymal stem/stromal cells (MSCs) exert potent immunomodulatory effects that are increasingly attributed not only to soluble factors but also to their extracellular matrix, membrane-associated cues and intracellular cargo. Consequently, MSC-derived devitalized biomaterials represent an attractive off-the-shelf strategy to deliver bioactive regulatory signals while avoiding the safety, storage and variability challenges associated with living cell therapies [1,2]. However, devitalization methods can profoundly alter cell architecture, organelles and ligand presentation, thereby reshaping both structural properties and immunological function. Here, we introduce high-pressure processing (HPP) as a high-potential technology for precise devitalization, with the ability to inactivate cells while better preserving phenotype, ultrastructure and organelles than conventional approaches. Yet, how HPP influences the capacity to tailor the immunomodulatory activity and structural properties of MSC-based materials, particularly in three-dimensional assemblies, remains poorly understood.

In this study, living, conventionally fixed and HPP-exposed MSC spheroids were compared as model cellular biomaterials with preserved multicellular organization. We assessed the impact of each condition on cell viability, spheroid integrity and structural features, and evaluated their ability to modulate inflammatory macrophage activation. Macrophages were stimulated with lipopolysaccharide (LPS) to activate the NF-κB pathway, and the regulatory effects of MSC spheroids were analysed through pathway activation readouts indicative of pro-inflammatory signalling.

By integrating viability analysis with functional macrophage assays, this work addresses a central gap in the development of MSC-derived devitalized biomaterials: whether preservation of cellular phenotype and organelle-rich architecture translates into retained or tailorable immunoregulatory function. The findings provide a basis to guide the rational selection of devitalization strategies for acellular immunomodulatory therapies and to determine whether HPP can generate structurally stable MSC assemblies capable of controlling inflammatory responses.

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P15: High hydrostatic pressure enhances cardiomyocyte contraction by regulating intracellular Ca^{2+} dynamics

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Cardiomyocytes are continuously exposed to haemodynamic stress, including hydrostatic pressure, which contributes to the regulation of cardiac function. In chronic hypertension, persistently elevated hydrostatic pressure induces cardiac hypertrophy and heart failure via impaired Ca^{2+} handling, suggesting that cardiomyocytes are highly sensitive to pressure changes. However, these findings mainly derive from *in vivo* and organ-level studies, and the acute, direct effects of hydrostatic pressure on individual cardiomyocytes remain poorly understood because real-time quantitative measurements under tightly controlled pressure conditions are lacking.

In our previous experiments, we demonstrated that ultra-high hydrostatic pressure induced a slow shortening in single mouse cardiomyocytes[1]. A conventional high-pressure microscope was used to observe cellular dynamics under hydrostatic pressure in the tens of MPa range (20 MPa ≈ 150,000 mmHg). However, this conventional system could not support high-sensitivity fluorescence imaging, because high-numerical- aperture objective lenses could not be used with the thick observation window of the high-pressure chamber[2]. Additionally, the chamber lacked electrodes for electrical stimulation because it was closed system with a thick, pressure-resistant windows. Thus, electrically stimulated contraction in adult ventricular cardiomyocytes could not be evaluated using this older system.

In the present study, we developed a high-pressure microscope that enables high-resolution, high-sensitivity real-time imaging with high-numerical-aperture objective lenses. Ventricular myocytes isolated from 129/Sv wild-type mice were stimulated at 1 Hz and exposed to 200 mmHg hydrostatic pressure in a high-pressure chamber filled with normal Tyrode solution. High hydrostatic pressure significantly increased twitch contraction (**Figure 1**) and Ca^{2+} transient amplitude compared with atmospheric conditions. These pressure-induced responses were abolished by both pharmacological inhibition and genetic deletion of transient receptor potential canonical 6 (TRPC6), a cardiac non-selective cation channel. Collectively, our findings demonstrate that the hydrostatic pressure at 200 mmHg enhances cardiomyocyte contraction via TRPC6-dependent regulation of intracellular Ca^{2+} dynamics, providing a cellular mechanism linking hydrostatic pressure overload to cardiac function.

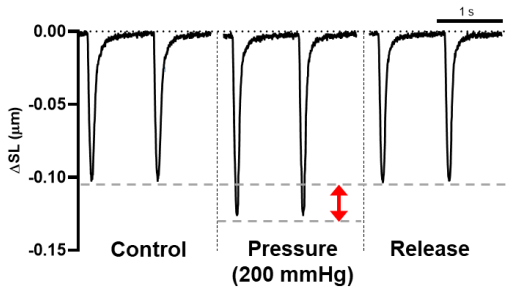

Figure 3. Effect of high hydrostatic pressure on twitch contraction in mouse ventricular cardiomyocytes. Applying a hydrostatic pressure of 200 mmHg increased the twitch contraction. This increase returned to the initial state when the pressure was released.

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Structure, Dynamics and Function of Biomacromolecules under High Pressure

O16: Insights into the Structure of Invisible Conformations of Large Methyl Group Labeled Molecular Machines from High Pressure NMR

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Most proteins are highly flexible and can adopt conformations that deviate from the energetically most favorable ground state. Structural information on these lowly populated, alternative conformations is often lacking, despite the functional importance of these states. Here, we study the pathway by which the Dcp1:Dcp2 mRNA decapping complex exchanges between an autoinhibited closed and an open conformation. We make use of methyl Carr-Purcell-Meiboom-Gill (NMR) relaxation dispersion (RD) experiments that report on the population of the sparsely populated open conformation as well as on the exchange rate between the two conformations.

To obtain volumetric information on the open conformation as well as on the transition state structure we made use of RD measurements at elevated pressures. We found that the open Dcp1:Dcp2 conformation has a lower molecular volume than the closed conformation and that the transition state is close in volume to the closed state. In the presence of ATP the volume change upon opening of the complex increases and the volume of the transition state lies in-between the volumes of the closed and open state.

These findings show that ATP has an effect on the volume changes that are associated with the opening-closing pathway of the complex. Our results highlight the strength of pressure dependent NMR methods to obtain insights into structural features of protein conformations that are not directly observable.

As our work makes use of methyl groups as NMR probes we conclude that the applied methodology is also applicable to high molecular weight complexes.

O17: Influence of high hydrostatic pressure pretreatment on the extractability and composition of citrus pectin

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Citrus processing generates large volumes of byproducts, with up to 50% of the fruit mass discarded as peel, representing a significant and underutilized source of pectin. Pectin is a structurally complex polysaccharide widely used in food and pharmaceutical applications due to its gelling, stabilizing, and functional properties.[1] In citrus peel, however, pectin exists as protopectin embedded within a highly organized matrix of cellulose and hemicellulose, where intermolecular interactions and physical entrapment limit its extractability and influence its final structural characteristics[2].

Conventional extraction methods, typically based on acid hydrolysis, are primarily optimized to maximize yield. Nevertheless, it is increasingly recognized that extraction conditions also affect pectin composition and molecular integrity, which ultimately determine its functional performance. As a result, maximizing recovery does not necessarily lead to improved material quality, highlighting the need to better understand the relationship between processing conditions and pectin structure[3].

In this context, non-thermal pre-treatment technologies such as high hydrostatic pressure (HHP) have been proposed as tools to modify plant cell wall architecture and enhance extraction processes. Pressure-induced structural changes may increase matrix accessibility and facilitate the release of pectic substances. However, pressure may also promote depolymerization or fragmentation of pectic chains, potentially reducing the amount of recoverable pectin or altering its composition[4,5]. These competing effects suggest that the influence of pressure on pectin extraction is not linear but rather governed by a balance between enhanced accessibility and structural degradation.

Orange peel was subjected to HHP at 100, 200, and 300 MPa under native (N) and hydrated (H) conditions during come-up time (CUT). Peel hardness was measured by texture profile analysis to assess matrix modification. The remaining material was dried, milled, and extracted using citric acid at pH 1.46, 80 °C for 30 min. Pectin yield was calculated on a dry basis (n = 4). Based on screening results, only the N series was further characterized. Galacturonic acid content (GalA)[6] was determined by the colorimetric carbazole method, and degree of esterification (DE) was estimated by Fourier-transform infrared spectroscopy (FTIR)[7].

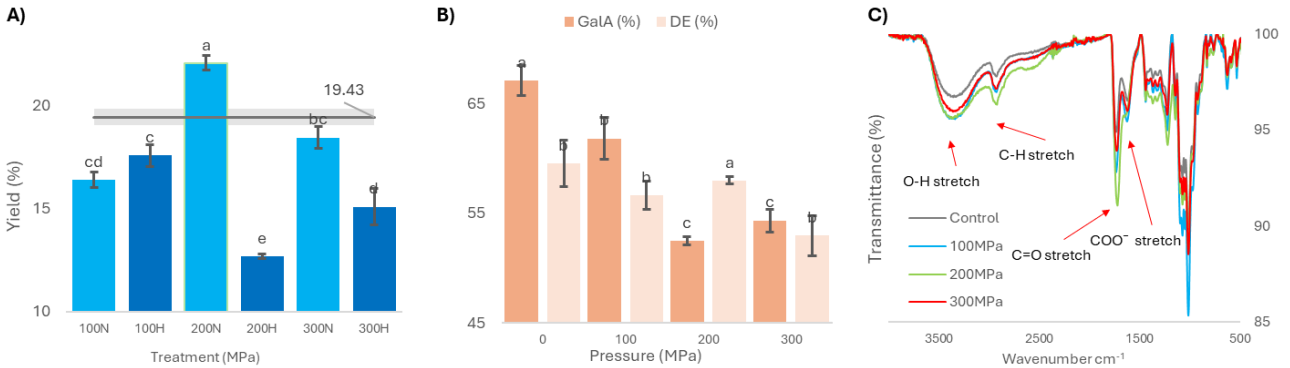

Figure 4. Effect of HHP on pectin yield, composition, and FTIR spectra. (A) Yield after HHP pretreatment at 100–300 MPa under native (N) and hydrated (H) conditions; the horizontal line represents control yield. (B) GalA and DE of control and native-pressure pectin samples. (C) FTIR spectra showing the main bands of pectin, including esterified carboxyl groups (~1740 cm^{-1}) and ionized carboxyl groups (~1600 cm^{-1}). Values are mean ± SD. Different letters indicate significant differences by one-way ANOVA and Tukey's test ($p < 0.05$). n = 4

HHP response depended on both pressure and hydration state (**Figure 1A**). Under native conditions, 200 MPa produced the highest yield (22.06 ± 0.10%), 13.5% above the control (19.43 ± 0.39%), whereas hydrated treatments reduced recovery, particularly 200H (12.68 ± 0.52%). Texture results supported this trend, since peel hardness was lowest for 200N (9083 ± 317 g) and highest for 200H (14964 ± 818 g), indicating contrasting degrees of matrix disruption. In the native series, pectin composition was also pressure-dependent (**Figure 1B**).

GalA decreased from 67.1% in the control to 52.4–54.3% at 200–300 MPa, while DE varied within a narrower range, with 200 MPa showing the highest value. FTIR spectra confirmed structural modifications after HHP (**Figure 1C**), particularly in the bands near ~1740 cm^{-1} and ~1600 cm^{-1}, associated with esterified and ionized carboxyl groups, respectively. Overall, 200 MPa under native conditions improved extractability, but this advantage was accompanied by compositional changes, especially lower GalA. Further studies on pressure-time interactions are needed to define conditions that maximize yield without compromising pectin integrity and functionality.

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O18: Adaptation to high salinity: Response of bacterial model membranes to Martian deep subsurface conditions

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In recent years, research on extremophiles has gained increasing importance due to their ability to thrive in extreme environmental conditions. Understanding the limits of life and its associated molecules from a physicochemical perspective is of great significance in biology and astrobiology.[1-3] Despite numerous examples of organisms thriving at high salt concentrations, high pressure, and extreme temperatures on Earth, there is little research on extraterrestrial environments that could support life[2]. Against this backdrop, and considering that membranes are essential for the development of all life forms, we focused our study on a bacterial model membrane in a potential Mars-like habitat to assess whether a microorganism could thrive in such an environment. Mars is believed to contain subsurface lakes with high concentrations of salts such as magnesium perchlorate, sulphate, and chloride.[2] We investigated the impact of these salts, in combination with temperature and pressure variation, on a model membrane composed of phosphatidylethanolamine and phosphatidylglycerol, using a range of biophysical techniques including calorimetry, spectroscopy, microscopy, and small-angle X-ray scattering. Our findings revealed that, despite very high salt concentrations, the lipids were still able to form a lipid bilayer membrane. Moreover, we found that the chaotropic perchlorate ion stabilizes the liquid-like bilayer phase that is necessary for a living cell to function, thereby counteracting the deteriorating effect of high pressure in the Martian subsurface. In contrast, high Mg^{2+} salt concentrations stabilize the rigid gel phase of the membrane, rendering the ability of bacteria to survive under such environmental conditions less favorable.[4]

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People and equipment behind high pressure research at University of Aveiro

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University of Aveiro high pressure equipment portfolio

Laboratorial scale	Pilot scale	Industrial scale

Multivessel pressure system 1	Multivessel pressure system 2

Connect with us!

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