

Development and technological functionality of micro and nano sized extracellular vesicles from plant matrices intended for food applications

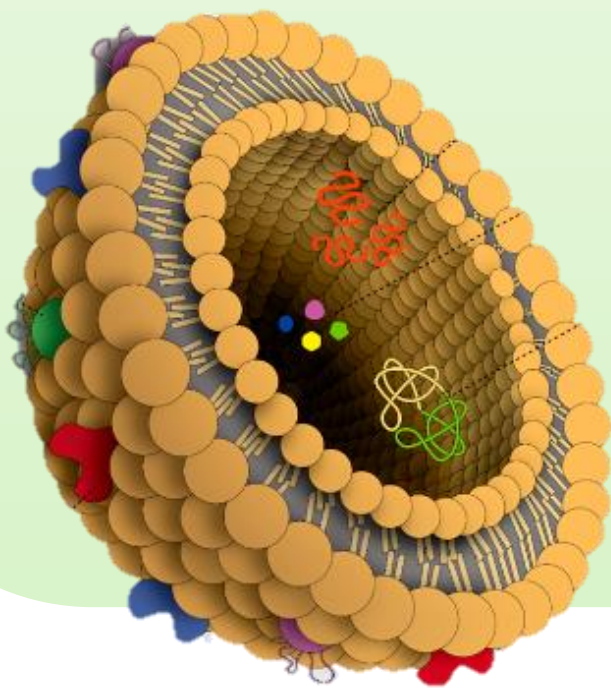
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Projects objective

This research project aims to develop innovative food ingredients with high bioactive potential by encapsulating extracellular vesicles from plant matrices and by-products. The vesicles will be characterized for their physical, technological, and bioactive properties. Using spray-drying with carriers such as maltodextrins and hydrocolloids, stable powdered ingredients will be produced. These encapsulated vesicles will then be tested for their effectiveness and performance in food products like sauces and cookies.

State of art

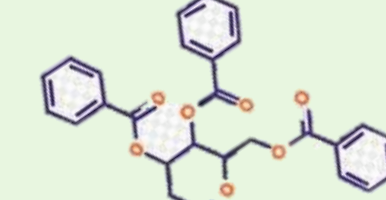
Extracellular vesicles (EVs) are lipid bilayer vesicles ranging from 50 to 5000 nm in size, produced by mammalian, plant, and bacterial cells. In the past, their function as delivery vectors for cellular waste was defined, while recent studies highlight their key role in cell-to-cell communication, transporting nucleic acids, proteins, and metabolites (Lo et al., 2024). EVs were formally defined by the International Society for Extracellular Vesicles (ISEV) in 2018 as "particles naturally released from cells that are delimited by a lipid bilayer and cannot replicate" (Pinedo et al., 2021). EVs are categorized into exosomes, microvesicles, and apoptotic bodies based on size and origin. Plant-derived extracellular vesicles (PDEVs) have gained attention for their role in cross-kingdom communication and potential health benefits. PDEVs consist of a phospholipid bilayer enclosing proteins and nucleic acids, with sizes ranging from 100 to 1000 nm and a negative zeta potential, depending on the plant species (Urzi et al., 2021). PDEVs from sources like citrus fruits, ginger, and garlic have shown potential in improving human health (Logozzi et al., 2022). PDEVs are effective carriers for natural bioactive compounds, enhancing their bioavailability and protecting them during transport to target cells. However, challenges such as cost-effective production and stability during storage remain. Encapsulation methods like freeze-drying and spray-drying show promise in creating stable, powder-like PDEV microcapsules, which could be applied in a variety of food products (Lian et al., 2022).

Objectives and Milestones

A1) Optimization of the extraction process of PDEV from different vegetables sources such as hops, tea leaves and grapefruit. Standardized methodology will be used for extraction and the yield of extraction evaluated based on the physical properties (number, size) of PDEV.



A2) Characterization of the physical, structural, technological functionality of selected PDEV. PDEV from one plant matrix will be selected and a full characterization will be performed on the physical (size, zeta-potential, Nanoparticle Tracking Analysis) bioactivity (DPPH and FRAP assays, antimicrobial testing) and content of bioactives via HPLC analysis.



A3) Encapsulation of PDEV. Two different technologies, freeze-drying and spray-drying will be tested by using different carriers like maltodextrin, arabic gum, and inulin and the corresponding microcapsules characterized for their chemical physical and technological performances, including moisture, solubility, and content of bioactives. Process encapsulation efficiency will be evaluated and the stability of the powders tested under varied stressing conditions (temperature, aw) conditions.

A4) Evaluation of encapsulated PDEV in model and real-like food matrices. The microencapsulated PDEV will be tested in model (gels at different aw) and real-like foods (e.g. mayonnaise and cookies) and their efficiency in terms of antioxidant and antimicrobial properties assessed. The real-like food matrices will be also evaluated for their overall quality (e.g. colour, colloidal properties), stability, and sensory properties over storage time.



A5) Writing and Editing of the PhD thesis, scientific papers and oral and/or poster communications

Activity	Months	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
A1) Extraction of PDEV																									
1) Tea, pomegranate, hops																									
A2) Characterization of PDEV																									
1) NTA, Zeta sizer																									
2) DPPH, FRAP, HPLC																									
A3) Encapsulation of vesicles																									
1) Spray dryer optimization																									
2) Characterization of powder																									
A4) Evaluation in food																									
1) Evaluation of quality and stability																									
2) Application in different food matrices																									
A5) Thesis and Paper Preparation																									

What for?



Development of high bioactivity food ingredients derived from plant-based extracellular vesicles, obtained by encapsulation techniques and intended to be applied in food products to enhance health benefits and stability

Selected References

- Logozzi M. et al, Int. J. Mol. Sci. 2022, 23, 4919. DOI: 10.3390/ijms23094919
- Kai-Jiun Lo et al., J. Agric. Food Chem. 2024 72 (6), 2853-2878 DOI:10.1021/acs.jafc.3c06867
- Pinedo M et al (2021) J. Extracell. Vesicles, 10: e12048. DOI:10.1002/jev2.12048
- Urzi O. et al (2021). Int. J Mol. Sci., 22(10), 5366.DOI:10.3390/ijms22105366
- Lian M. et al (2022). J. Extracell. Vesicles, 11(12), e12283. DOI:10.1002/jev2.12283