Whey protein-based hydrogel as phenylethyl isothiocyanate delivery system

Ana Sofia Sousa[1], Ezequiel R. Coscueta[1], Celso A. Reis[2,3,4], Manuela Pintado[1]*

[1]Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Rua Diogo Botelho 1327, 4169-005 Porto, Portugal; *mpintado@ucp.pt.

[2]i3S – Instituto de Investigação e Inovação em Saúde, University of Porto, 4169-005 Porto, Portugal; celsor@ipatimup.pt

[3]IPATIMUP – Institute of Molecular Pathology and Immunology, University of Porto, 4169-005 Porto, Portugal

[4]Medical Faculty, University of Porto, Al. Prof. Hernâni Monteiro, 4169-005 Porto, Portugal

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Introduction

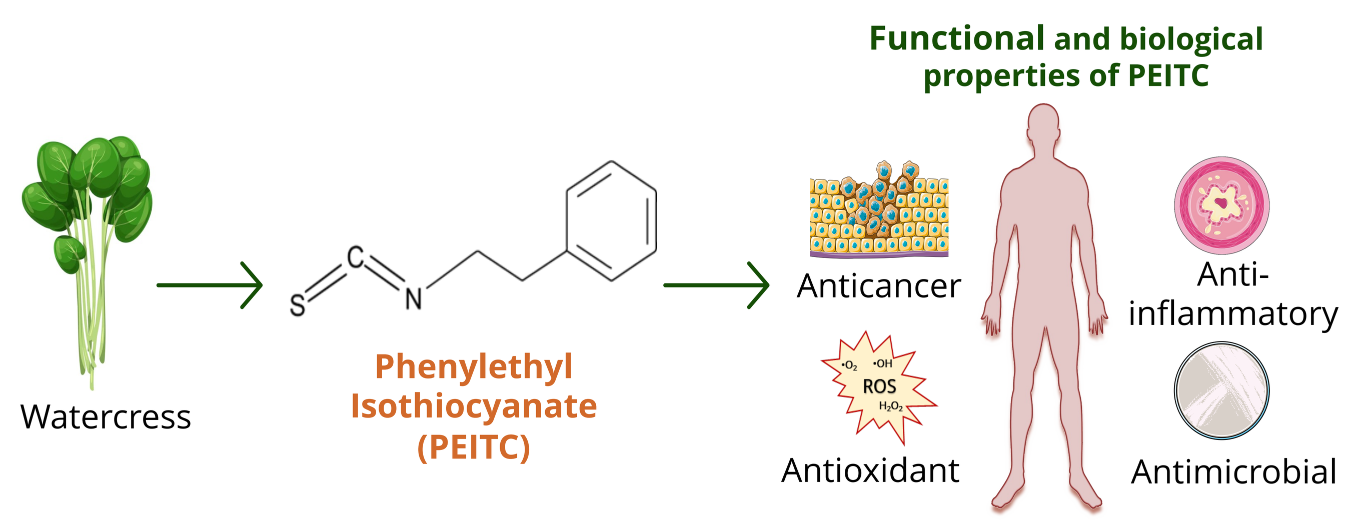

Figure 1 - The main effects of PEITC on human health.

Although PEITC is a promising health-promoting compound, its industrialisation has been complex due to its relative instability and reactivity. In this sense, the encapsulation of this agent can solve these limitations. On the other hand, protein-based hydrogels have received considerable attention because of their conventional use in food matrices and as promising bioactive components and nutraceuticals delivery systems. This type of hydrogel has outstanding properties, such as high nutritional value, excellent functional properties, amphiphilic nature, biocompatibility, biodegradability and lower toxicity compared to synthetic polymers. Among food-grade polymers, whey protein isolate (WPI) has gained interest and wide acceptance as a usual gelling agent for developing hydrogel formulations.

The MAIN GOAL of this study was to develop WPI hydrogel gelled via citric acid-mediated cross-linking as a potential delivery system for the PEITC.

Methods

Protein hydrogel was produced from a heat-denatured protein solution and subsequent cross-linking by citric acid, resulting in a cold-setting hydrogel. Then, PEITC, watercress extract (WE) and microparticles (MPs) loaded with PEITC or WE, previous developed using chitosan-olive oil systems, were added to the protein solution.

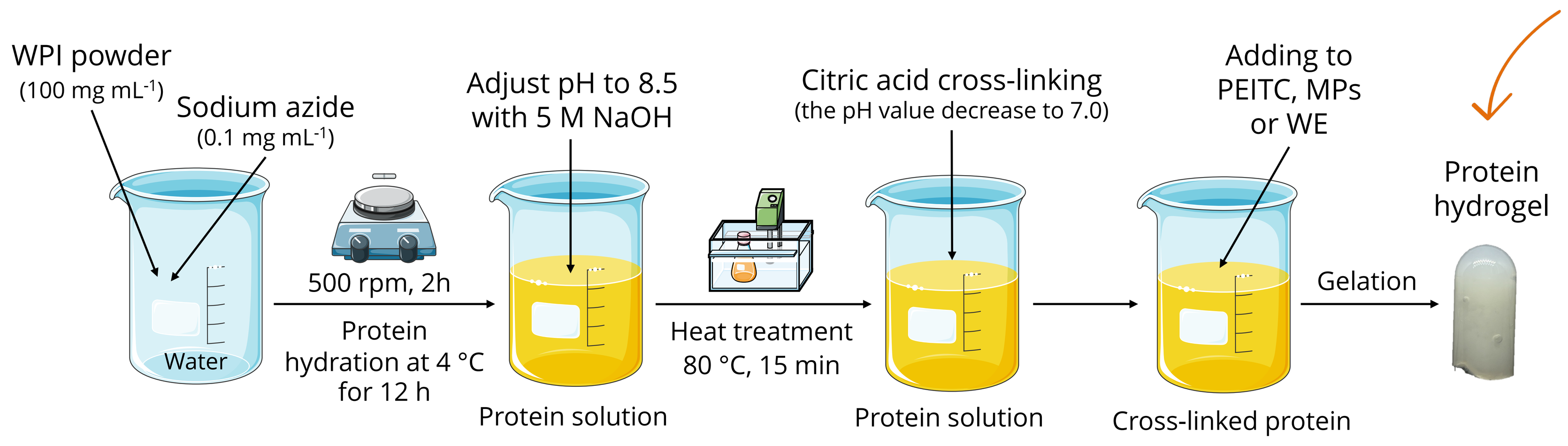

Figure 2 – Schematic illustration of the procedure of WPI gel formation

Table 2 - The composition of the proteins hydrogels

Whey protein isolate hydrogels							
Sample	Ingredients (%)						
	WPI solution	MP-Free	PEITC	MP + PEITC	WE	MP + WE	Water
WPI Hydrogel	75	-	-	-	-	-	25
WPI + MP-Free	75	0,33	-	-	-	-	24,67
WPI + PEITC	75	-	0,33	-	-	-	24,67
WPI + MP-PEITC	75	-	-	0,33	-	-	24,67
WPI + WE	75	-	-	-	11,77	-	13,23
WPI + MP-WE	75	-	-	-	-	25	-

ANALYSIS

- ✓ Rheological analysis
- ✓ Scanning electron microscopy (SEM)
- ✓ Water-holding capacity (WHC)
- ✓ Fourier transform infrared (FTIR) spectroscopy
- ✓ *In vitro* gastric digestibility
- ✓ Gel swelling

STATISTICAL ANALYSIS

- Anova, Tukey test ($p < 0.05$)

Results

WPI hydrogels

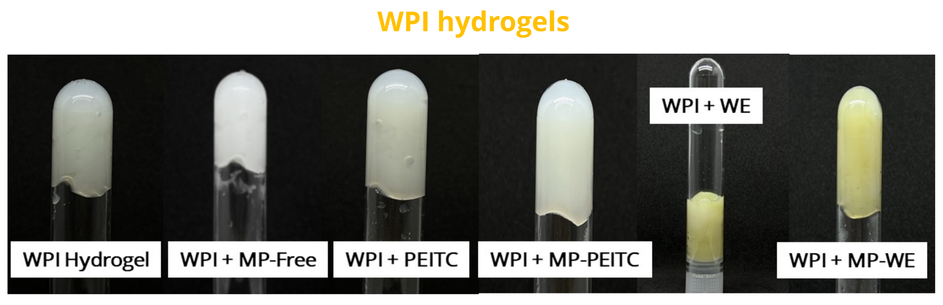

Figure 3 – Schematic illustration of the procedure of WPI gel formation

The WPI hydrogel formulation showed adequate structural integrity, with no visible pores or cracks.

SEM

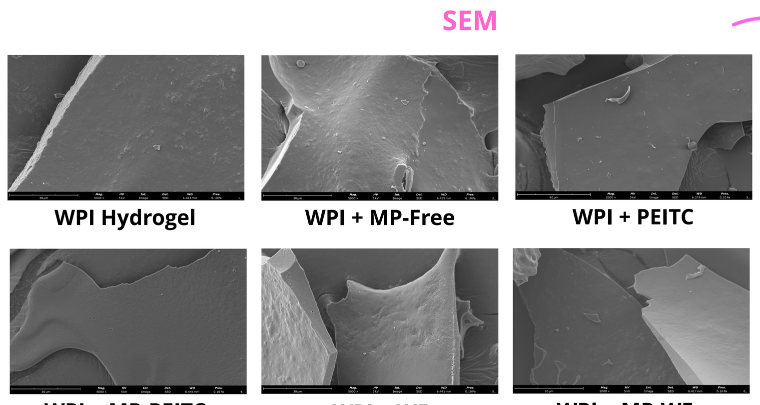

Figure 4 - SEM micrographs of WPI hydrogels.

The SEM results showed that the surface appears relatively smooth.

WHC

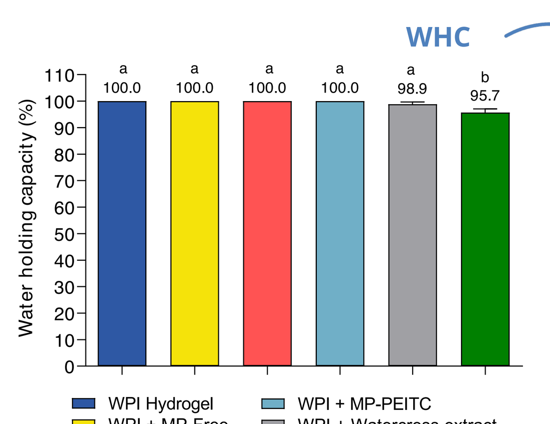

Figure 7 - WHC of WPI hydrogels. The values are expressed as mean ± standard deviation (n=3). Values with different letters indicate significant differences between each mean ($p < 0.05$).

The gels showed a good ability to retain water, which is desirable as water loss can cause gel shrinkage, texture change and poor-quality attributes.

Rheological analysis

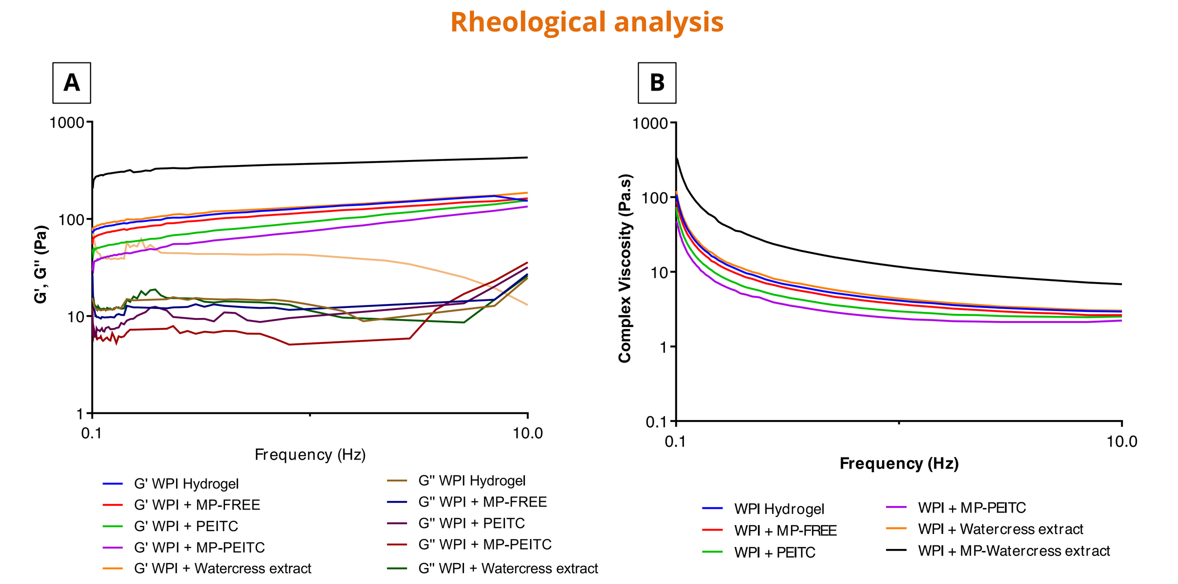

Figure 5 - The oscillatory scans displaying both the elastic modulus (G') and viscous modulus (G'') (A) and complex viscosity of WPI gels (B). The values are expressed as a mean (n=3).

The results showed that the G' > G'' indicated that the samples exhibited distinct elastic behaviour, usually characteristic of hydrogels. The gels with WE showed the highest G' values, making the gel less compact. The viscosity of all hydrogels decreased with increasing frequency rate.

FTIR spectra

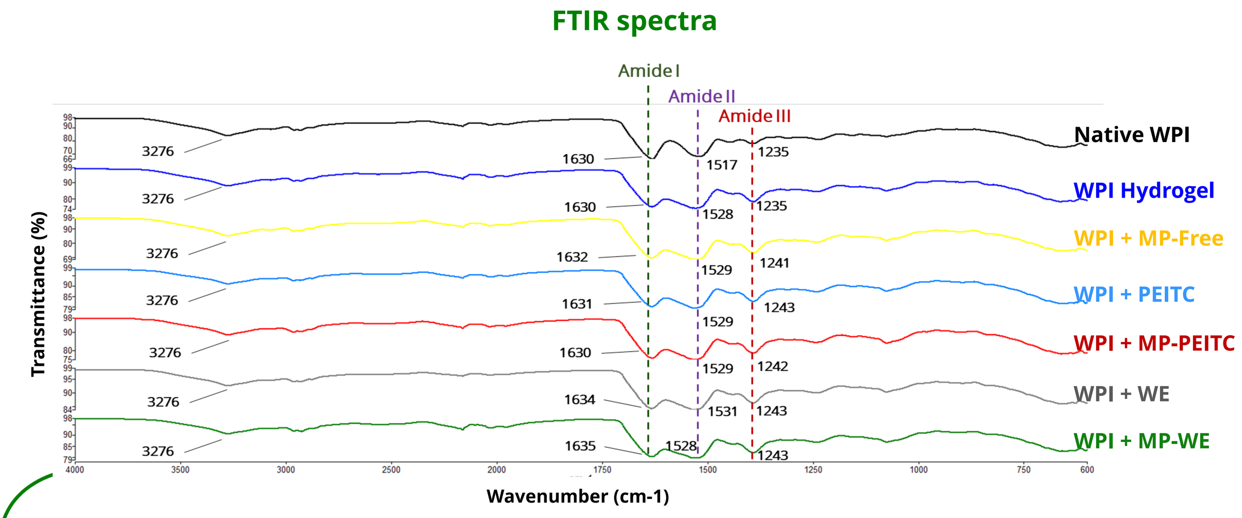

Figure 8 - The FTIR spectra of WPI hydrogels.

FTIR results showed that citric acid cross-linking did not produce disordered whey proteins. Indeed, the spectra of hydrogel samples are similar to each other.

In vitro gastric digestibility in the absence and presence of pepsin

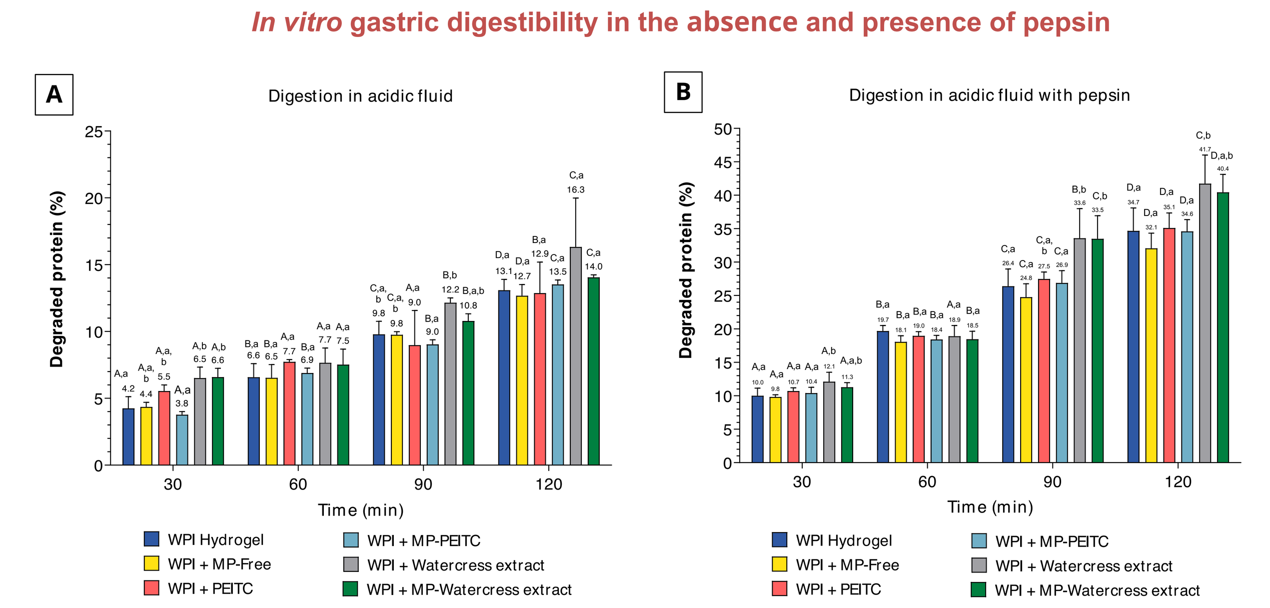

Figure 6 - Digestibility of WPI hydrogels in acidic fluid in the absence (A) and presence of pepsin (B). The results are expressed as mean ± standard deviation (n=4). Different letters indicate significant differences ($p < 0.05$). The capital letters indicate the differences among the time in the same gel, and small letters indicate the differences between distinct hydrogels in the same time.

The pepsin digestion assay resulted in much higher digestibility, increasing degraded protein for all samples over time.

Gel swelling

Table 2 - Swelling of WPI hydrogel samples.

Sample	Swelling (%) 30 min	60 min	90 min	120 min
WPI Hydrogel	6.2 ± 1.6 ^{A,a}	17.6 ± 3.1 ^{A,a}	19.6 ± 2.5 ^{B,a}	22.9 ± 1.4 ^{B,a}
WPI + MP-Free	4.6 ± 0.4 ^{A,a}	13.4 ± 1.8 ^{B,a}	15.8 ± 1.6 ^{B,a}	18 ± 2.1 ^{B,a}
WPI + PEITC	9.0 ± 1.6 ^{A,a}	15.6 ± 2.5 ^{A,a}	19.2 ± 3.2 ^{B,a}	22.1 ± 2.7 ^{B,a}
WPI + MP-PEITC	8.0 ± 1.9 ^{A,a}	13.5 ± 1.9 ^{B,a}	18.2 ± 2.8 ^{C,a}	22.3 ± 2.4 ^{D,a}
WPI + WE	13.4 ± 1.0 ^{A,a}	19.1 ± 1.3 ^{A,B,a}	22.0 ± 0.8 ^{B,C,a}	24.4 ± 1.4 ^{C,a}
WPI + MP-WE	14.7 ± 1.7 ^{A,a}	15.4 ± 1.5 ^{A,a}	18.3 ± 1 ^{A,a}	22.9 ± 1.3 ^{B,a}

The results are expressed as mean ± standard deviation (n=3). Different letters indicate significant differences. The capital letters indicate the differences among the time (30, 60, 90 and 120 min) in the same gel, and small letters indicate the differences between distinct hydrogel samples in the same point time.

All samples swelled in quiescent immersion in an acidic solution, significantly increased over time.

Conclusions

The results indicated that the addition of phenylethyl isothiocyanate, pure or encapsulated, did not affect the structure and characteristics of the gel. Therefore, the optimised protein hydrogel is an effective delivery system to encapsulate and release PEITC with potential applications in functional foods.

Acknowledgements

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