

Watercress extract: antioxidant potential and impact of different drying methods

Ana Sofia Sousa^{1*}, Ezequiel R. Coscueta¹, Celso A. Reis^{2,3,4}, Manuela Pintado¹

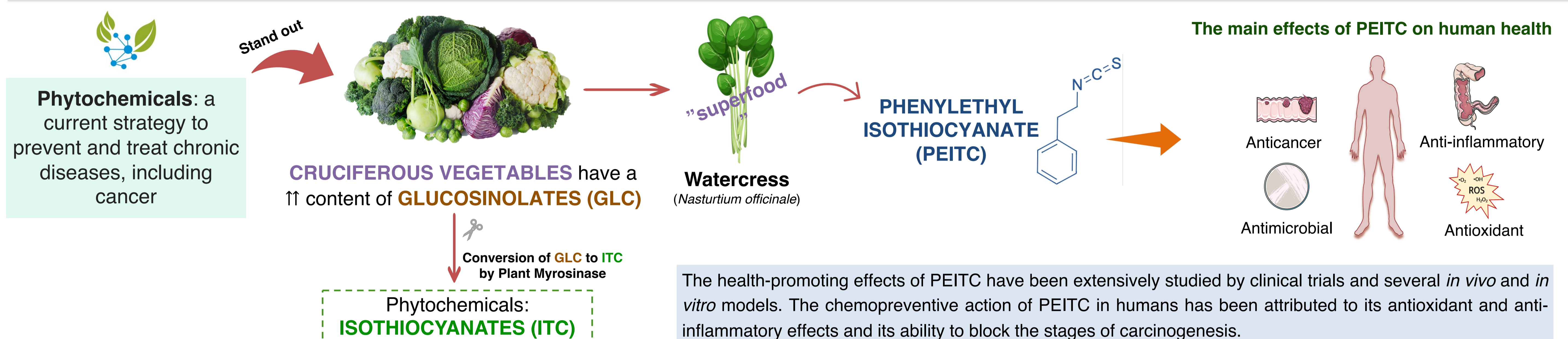
¹Universidade Católica Portuguesa, CBQF - Centro de Biotecnologia e Química Fina – Laboratório Associado, Escola Superior de Biotecnologia, Rua Diogo Botelho 1327, 4169-005 Porto, Portugal; *assousa@ucp.pt.

²i3S – Instituto de Investigação e Inovação em Saúde, University of Porto, 4169-005 Porto, Portugal;

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

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

Introduction & Objectives



The **MAIN GOAL** of this work is to evaluate the antioxidant potential and biocompatibility of a watercress extract obtained by a green extraction technique. Furthermore, in order to obtain a dry product, different drying techniques were tested, as well as their impact on the compound of interest (PEITC). Finally, polymeric microparticles were developed using the electrospraying technique to develop food-grade delivery vehicles for PEITC extracts.

PEITC was obtained in a circular economy context by valorising watercress by-products.

Methods & Results

Watercress by-products were provided by Vitacress Portugal S.A. The extraction was performed using a green technique.

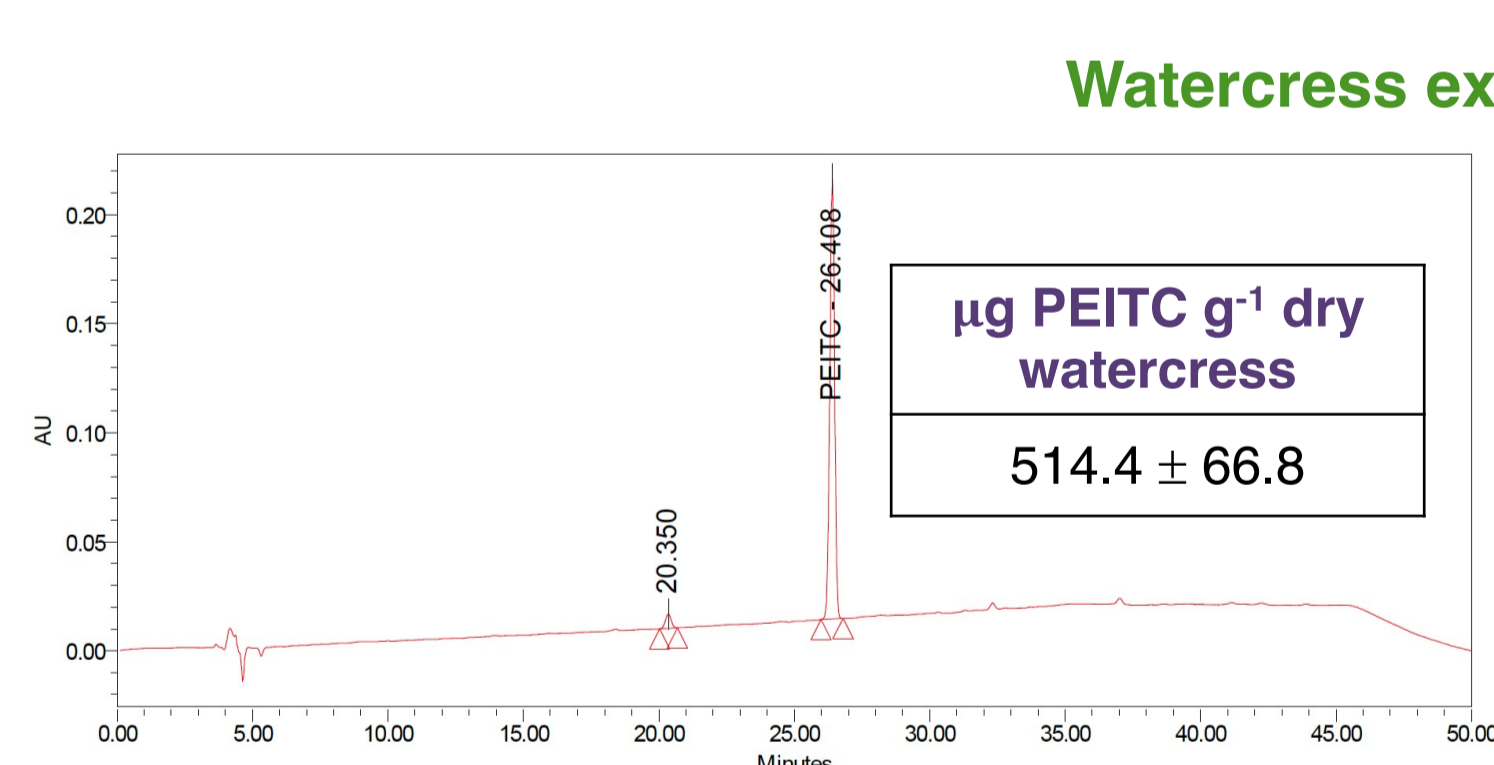
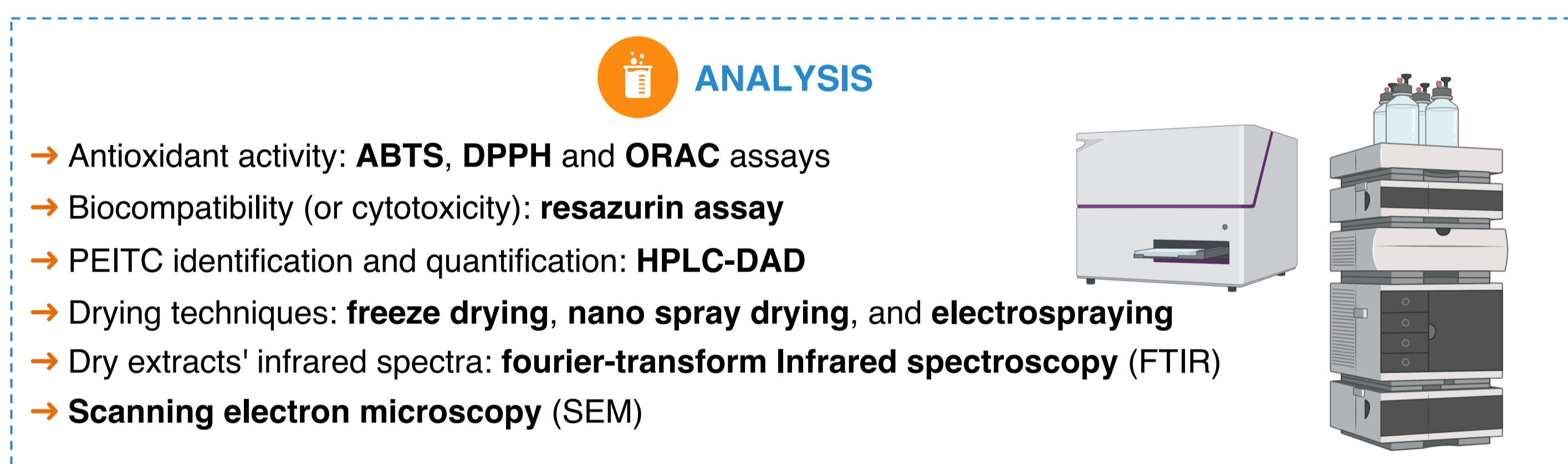


Figure 1. A typical HPLC chromatogram of PEITC identified in watercress extract at a wavelength of 245 nm.

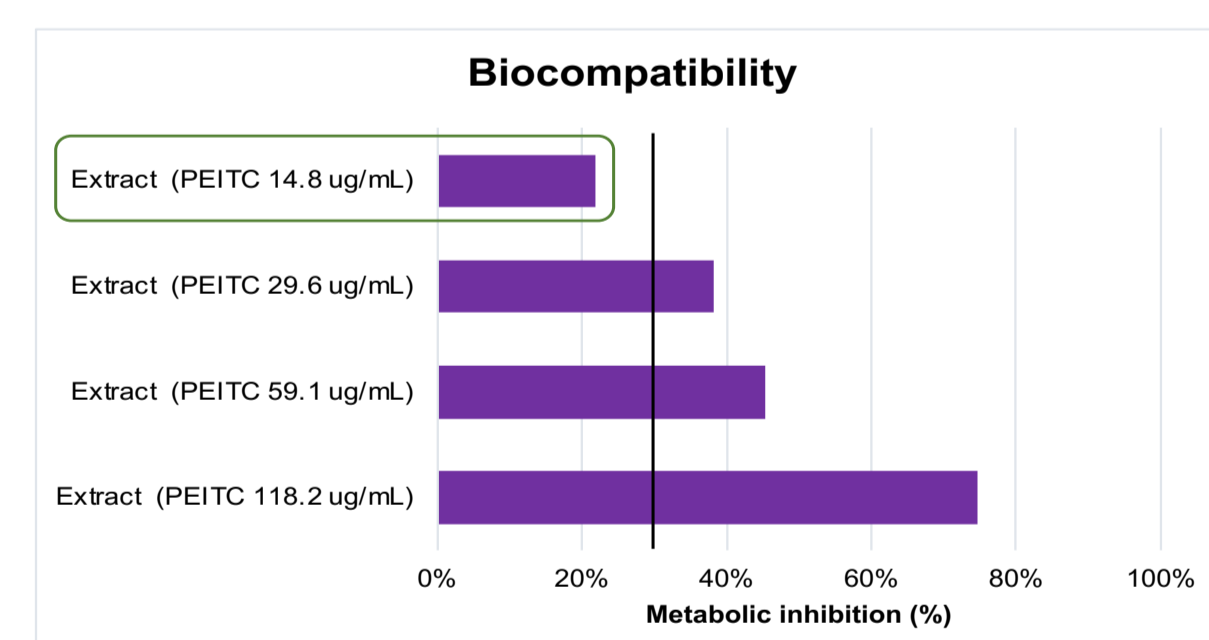


Figure 2. Metabolic inhibition of watercress extract at different equivalent concentrations of PEITC for 24h against Caco-2 cells. The black vertical line represents the 30% inhibition limit.

Table 1. Antioxidant activity determined by ABTS and DPPH assays of watercress extract.

	ABTS			DPPH		
	Linear equation	R ²	EC50 (µg/ml)	Linear equation	R ²	EC50 (µg/ml)
Trolox (control)	y = 8.2256x + 0.0571	0.999	53.8	y = 20.687x - 0.062	0.995	27.1
Watercress extract	y = 0.5144x + 0.076	0.994	824.3	y = 0.5746x + 0.1412	0.983	624.4

EC 50: the effective sample concentration that can decrease the concentration of ABTS or DPPH by 50%.

Regarding the EC₅₀, the results demonstrated that the extract was less effective for scavenging than trolox; Moreover, according to the international standard, ISO 10993-5, the threshold value for a sample to be cytotoxic is a metabolic inhibition of 30%. It is possible to observe that PEITC below 14.8 µg/mL slightly inhibited cell metabolism, corresponding to 21.78% ± 0.10%, but did not show cytotoxicity.

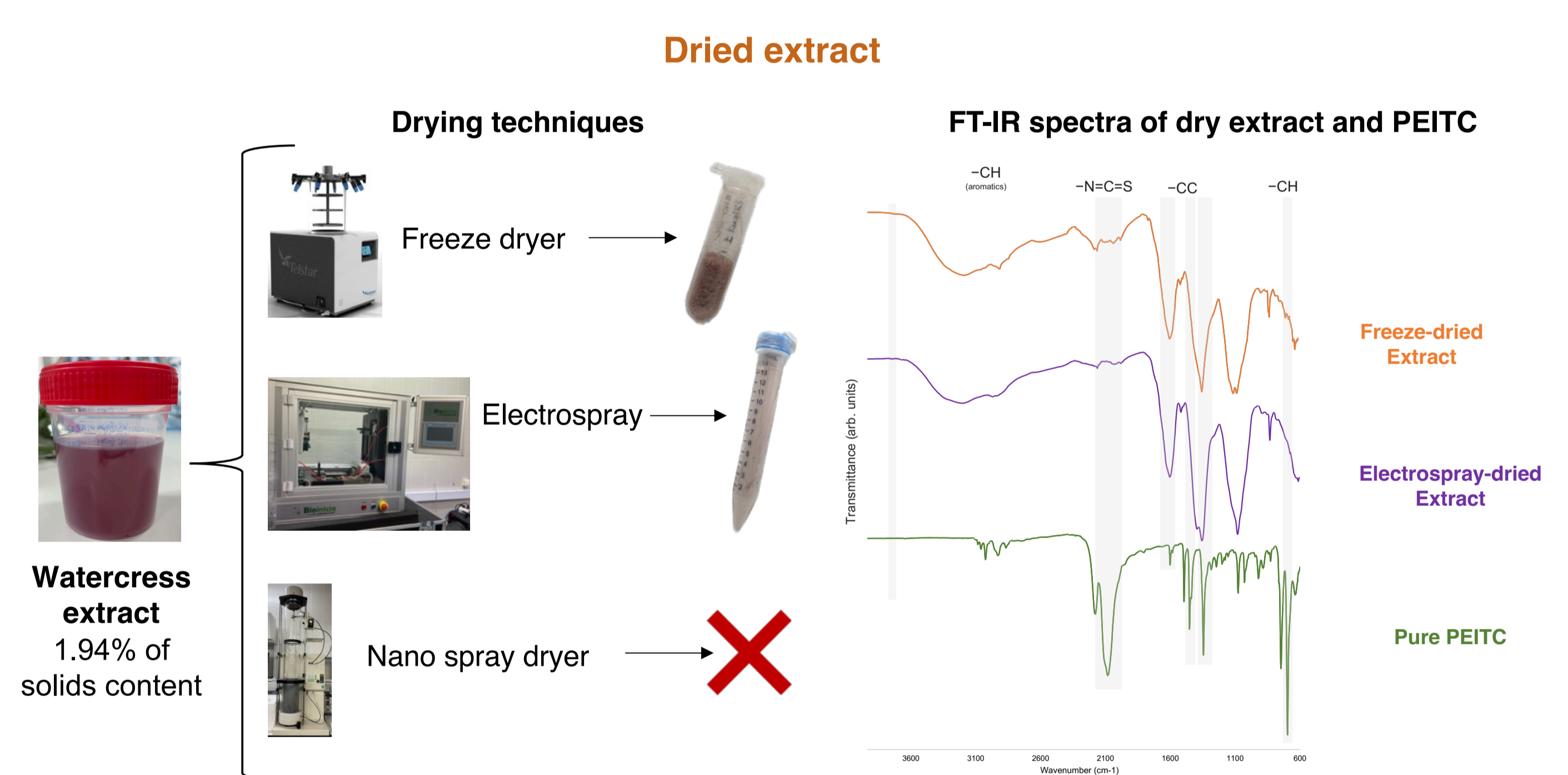


Figure 3. Dried watercress extract obtained by freeze dryer, electrospray, and FTIR analysis of the respective samples.

Regarding drying techniques, nano spray drying was not the best to obtain a dry extract. Drying the extract with a 1% maltodextrin solution was also tested, but it was not very effective in getting a powder. Besides, nano spray drying works with high temperatures, which is a disadvantage since some substances are thermosensitive, such as phenolic compounds. Electrospraying allowed for obtaining a powder with small and homogeneous particles. Figure 1 also shows pure PEITC and dry extracts' infrared spectra (FTIR). In the samples, it is possible to observe characteristic peaks of PEITC, such as the characteristic peak of the isosulfonyl group (N=C=S) between 2182 cm⁻¹ and 2083 cm⁻¹.

Table 2. Antioxidant activity determined by ORAC assay of dried extracts.

	Freeze-dried extract	Electrosprayed extract
µmol trolox equivalent / g dry extract	341 ± 39	499 ± 34

The electrosprayed extract showed a higher antioxidant potential. Furthermore, polymeric microparticles were developed and chitosan was chosen as encapsulation matrices as biocompatible, biodegradable, and gastro-resistant. The optimal chitosan concentration for electrospraying conditions was 4% (w/v). It was found that samples with chitosan at >4% (w/v) tend to start forming nanofibers in addition to microparticles.

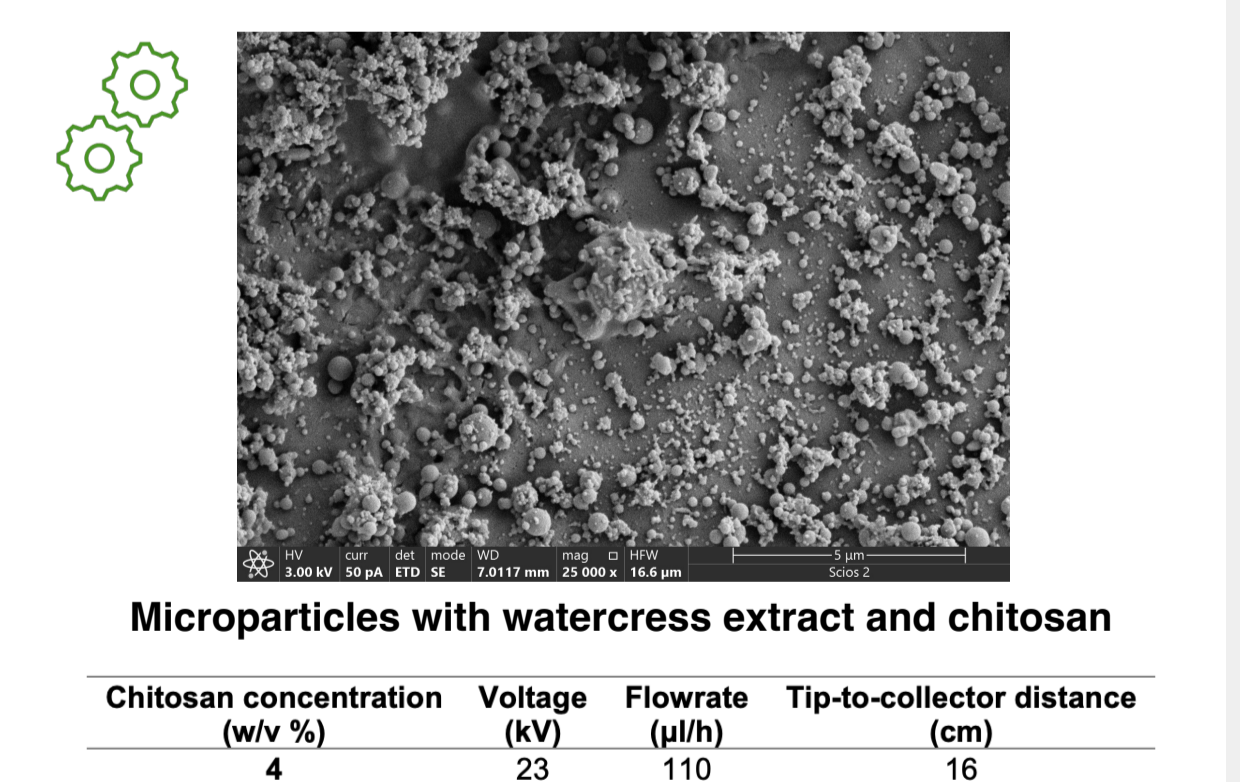


Figure 4. SEM image of the electrosprayed chitosan microparticles.

Conclusions & Future Work

According to the results, the obtained watercress extract is a promising source of PEITC. Regarding drying techniques, electrospray allows for obtaining a more homogeneous powder than the freeze-drying technique, with higher antioxidant activity. Further, the preliminary work in producing watercress extract microparticles showed potential and promising results, namely in producing microparticles on a pilot scale. Furthermore, future research will be carried out to characterise the dried extracts and the microparticles.

References

- Coscueta, E. R., Sousa, A. S., Reis, C. A. & Pintado, M. M. Phenylethyl Isothiocyanate: A Bioactive Agent for Gastrointestinal Health. *Molecules* **27**, 1–12 (2022)
- Coscueta, E. R., Sousa, A. S., Reis, C. A. & Pintado, M. Chitosan-olive oil microparticles for phenylethyl isothiocyanate delivery: optimal formulation. *PLoS One* **16**, 1–21 (2021)
- Gómez-Mascaraque, L. G., Sanchez, G. & López-Rubio, A. Impact of molecular weight on the formation of electrosprayed chitosan microcapsules as delivery vehicles for bioactive compounds. *Carbohydr Polym* **150**, (2016).

Acknowledgements

This work was supported by National Funds from FCT - Fundação para a Ciência e Tecnologia through project UIDB/50016/2020, author Ana Sofia Sousa individual PhD grant number 2021.07407.BD and through grant Ref.ª 2022.02926.PTDC "Nutraceutical biopolymeric-biocatalytic microbot against gut inflammatory disorders". The authors would also like to thank Vitacress Portugal S.A. for providing raw materials.