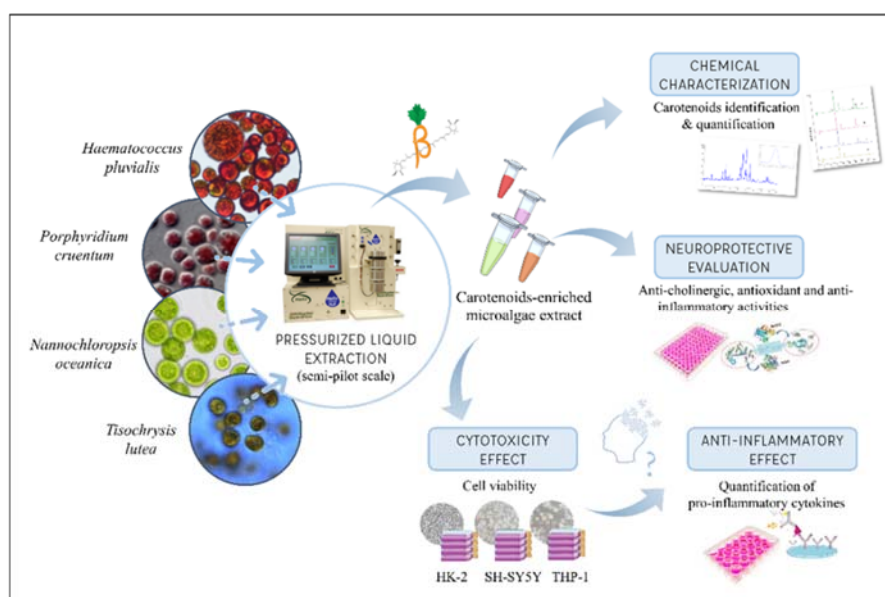


## Pressurized liquids extraction for obtaining microalgae extracts enriched in carotenoids with anti-inflammatory and neuroprotective effects

Rocío Gallego<sup>a</sup>, Alberto Valdés<sup>a</sup>, Zully J. Suárez-Montenegro<sup>a</sup>, José David Sánchez-Martínez<sup>a</sup>, Alejandro Cifuentes<sup>a</sup>, Elena Ibáñez<sup>a</sup>, Miguel Herrero<sup>a,\*</sup>

<sup>a</sup> *Laboratory of Foodomics, Institute of Food Science Research (CIAL, CSIC-UAM), Nicolás Cabrera 9, 28049 - Madrid, Spain*  
[m.herrero@csic.es](mailto:m.herrero@csic.es)

### GRAPHICAL ABSTRACT



### ABSTRACT

Microalgae are considered as an untapped reservoir for potential functional ingredients and high added-value compounds with diverse application in cosmetics, pharmaceutical and food industries [1]. Some of these bioactive compounds include polyunsaturated fatty acids, peptides, polyphenols, phytosterols or carotenoids [2]. Regarding carotenoids, these lipophilic compounds have been associated with an extensive list of health-promoting effects, including anti-inflammatory, neuroprotective or antioxidant properties [3], which emphasizes their potential value. In this study, the recovery of carotenoids from diverse microalgae, including extracts from *Haematococcus pluvialis*, *Nannochloropsis oceanica*, *Tisochrysis lutea* and *Porphyridium cruentum*, was performed using pressurized liquid extraction at semi-pilot scale, using the same optimum extraction conditions as obtained at lab-scale [4-7]. The extracts were chemically characterized by reversed-phase high-performance liquid chromatography with diode array detection (RP-HPLC-DAD) and then, these were evaluated through a battery of *in vitro* neuroprotective assays in an effort to estimate their potential against neurological disorders. Results indicated that microalgae extracts (obtained at semi-pilot scale) had

similar carotenoid profiles compared to extracts obtained at lab-scale, although higher yields were achieved due to the additional extraction cycle. All microalgae exerted a moderate and selective cholinesterase inhibitory potential, as well as high antioxidant and anti-inflammatory capacities, highlighting *N. oceanica* and *T. lutea* extracts. In parallel, cytotoxicity tests of the microalgae extracts were performed in different cell culture models, together with an *in vitro* evaluation of their anti-inflammatory capacity in THP-1 cells. In this regard, *N. oceanica* extract showed the highest inhibition of pro-inflammatory cytokine release, indicating that this microalga extract could be the most promising neuroprotective agent.

## ACKNOWLEDGEMENTS

This work was supported by projects ABACUS (Algae for a Biomass Applied to the production of added value compounds—funded by the Bio Based Industries Joint Undertaking under the European Union’s Horizon 2020 research and innovation programme under grant agreement No 745668) and AGL2017-89417-R (MINECO, Spain). The authors thank Microphyt (Baillargues, France) and Algae for Future (A4F, Lisbon, Portugal) for kindly providing microalgae biomasses. A.V. would like to acknowledge the Spanish Ministry of Science, Innovation and Universities for his “Juan de la Cierva” post-doctoral grant (IJC2018-037560-I). J.D.S-M. would like acknowledge the Ministry of Education for a FPU predoctoral grant FPU17/01876. Z.J.S-M. would like acknowledge the University of Nariño (Colombia) for financial support.

## REFERENCES

- [1] M. Bueno, C. Vitali, J.D. Sánchez-Martínez, J.A. Mendiola, A. Cifuentes, E. Ibáñez, M. Herrero. ACS Sustainable Chemistry & Engineering, 8(30), 11413–11423, 2020.
- [2] S.M. Cardoso, O.R. Pereira, A.M.L. Seca, D.C.G.A Pinto, A.M.S. Silva. Marine Drugs, 13(11), 6838–6865, 2015. <https://doi.org/10.3390/md13116838>.
- [3] R. Sathasivam, J.-S. Ki. Marine Drugs, 16(1), 26, 2018.
- [4] R. Gallego, K. Arena, P. Dugo, L. Mondello, E. Ibáñez, M. Herrero. Analytical and Bioanalytical Chemistry, 412, 589-599, 2020.
- [5] R. Gallego, M. Bueno, A.M. Chourio, E. Ibáñez, M.D.A Saldaña, M. Herrero. The Journal of Supercritical Fluids, 167, 105039, 2021.
- [6] R. Gallego, C. Tardif, C. Parreira, T. Guerra, M.J. Alves, E. Ibáñez, M. Herrero. Journal of Separation Science, 43, 1967–1977, 2020.
- [7] R. Gallego, M. Martínez, A. Cifuentes, E. Ibáñez, M. Herrero. Molecules, 24(8), 1564, 2019.