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Evaluation of *Chlorella vulgaris* extracts for skincare products

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Microalgae as Cosmetic actives for Skin Photoprotection

Photoaging, caused by UVA (320–400 nm) and blue light (420–500 nm), accelerates skin aging by generating reactive oxygen species (ROS), leading to oxidative stress. Active ingredients that neutralize ROS or absorb harmful radiation are key in skin protection. Microalgae, rich in **carotenoids**, offer a promising solution for cosmetics by preventing ROS formation and absorbing damaging rays. This study focuses on ***Chlorella vulgaris* strain CCALA924**, examining its pigment profile, antioxidant activity, and phenolic content. Special attention is given to optimizing cell wall disruption to improve the extraction of bioactive compounds for use in cosmetic products.



Origin of the molecules of interest



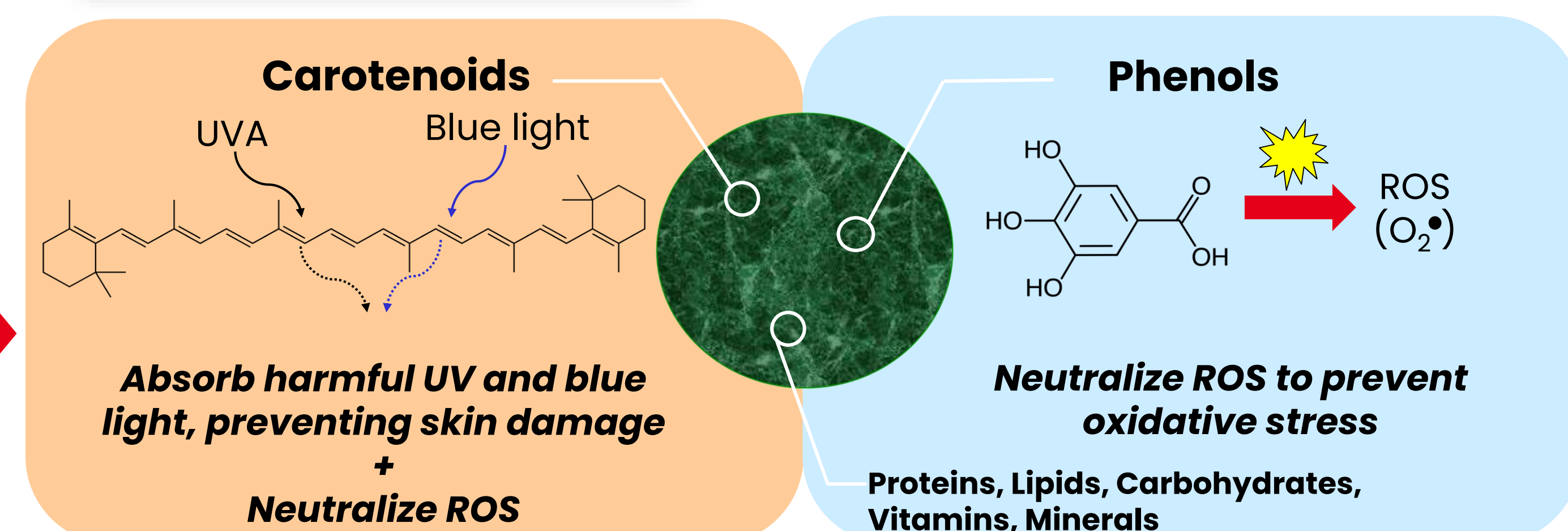
Growth conditions of *C.vulgaris* CCALA924

in Beijerinck medium :

- 2% CO₂; pH ~ 7.5

- 25°C

- 200 μE.m⁻².s⁻¹; 20:4 Day:Night (h)



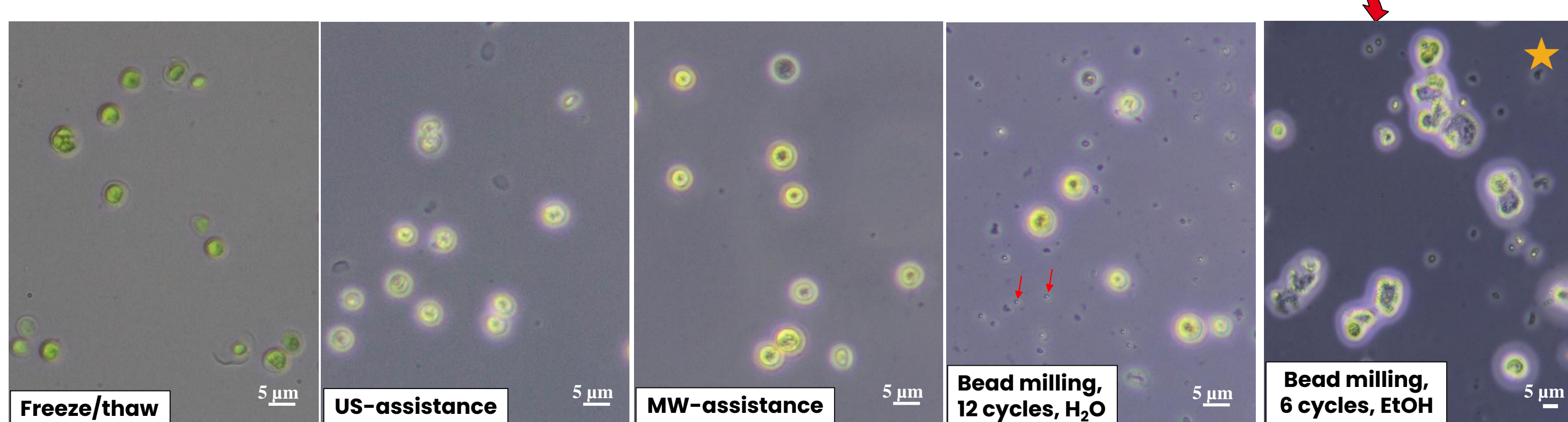
Cell wall disruption and Extraction

Materials and methods

- Wet biomass
- Solvent: EtOH 100%
- MM400 Retsch bead miller : glass beads (Ø=2mm), 30 Hz

Process	Time	Temperature (°C)
Maceration (M)	5 min	Ambient 25°C (Tamb)
	40 min	
	72 h	
Microwave (MW)	40 min	60°C
Ultrasound (US)	2 min	Tamb
★ Bead milling (B)	2 to 12 cycles of 2 min	Tamb

Cell wall disruption achieved



Microscopic Observations (400X) of *C.vulgaris* after various pre-treatments, from left to right : Freeze/thaw, Bead milling in H₂O cycle 12, US, MW, Bead milling in EtOH cycle 6

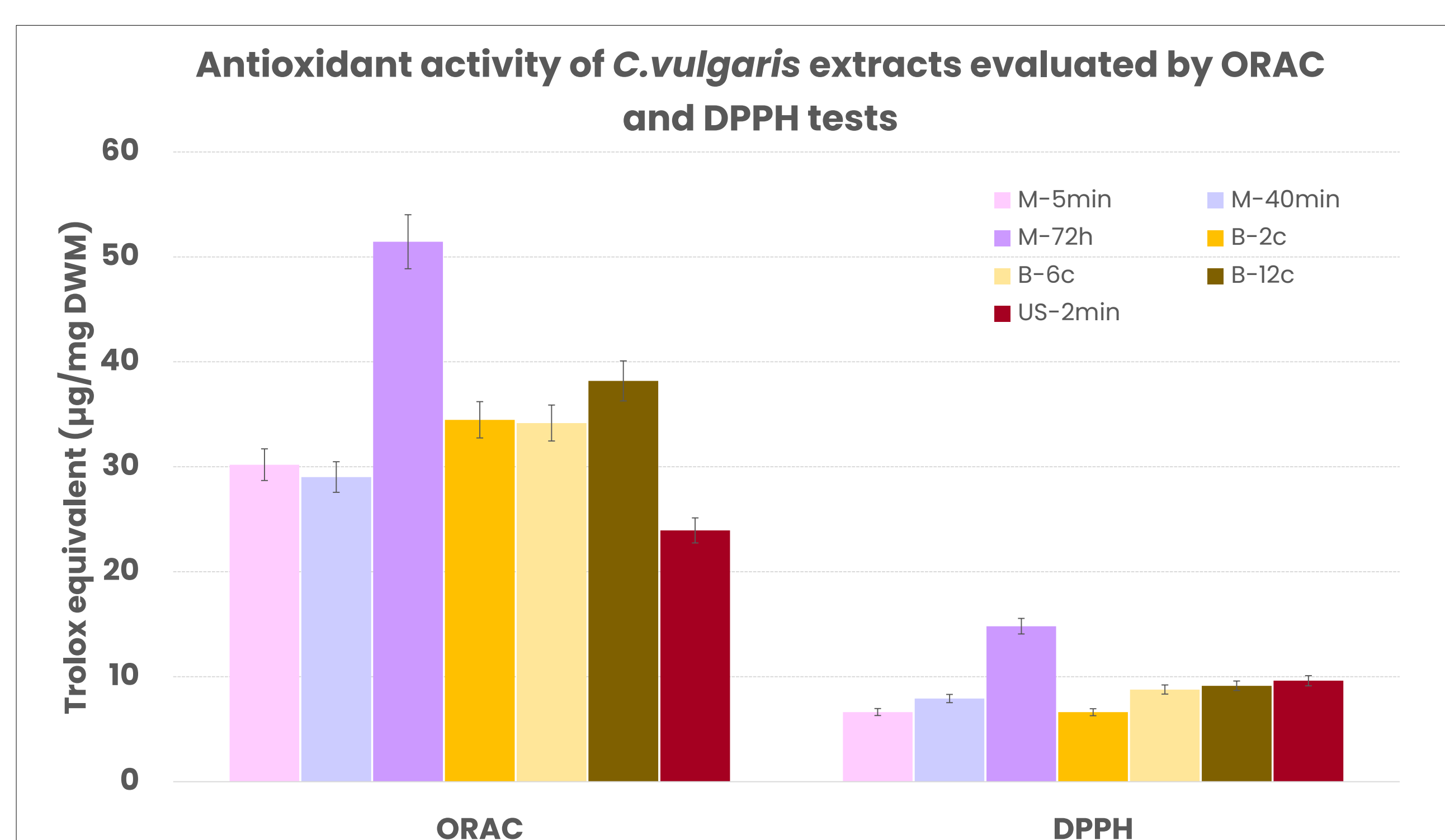
Antioxidant properties

Materials and methods

Two complementary antioxidant tests (Huang *and al.*, 2005; Brand-Williams *and al.*, 1995):

- ORAC, Spectrofluorimetry → reduction of peroxy radicals ROO•
- DPPH, Spectrophotometry → reduction of the stable DPPH radical

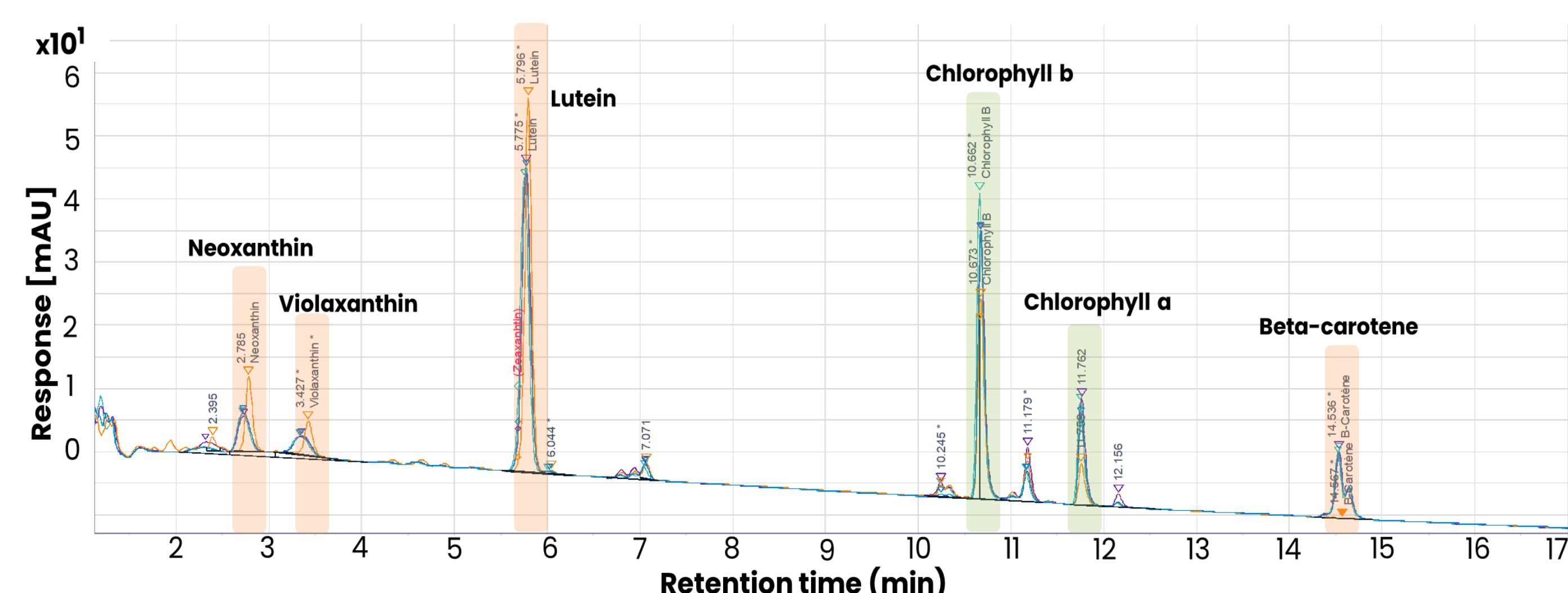
Standard : Trolox, equivalent of hydrosoluble Vitamin E



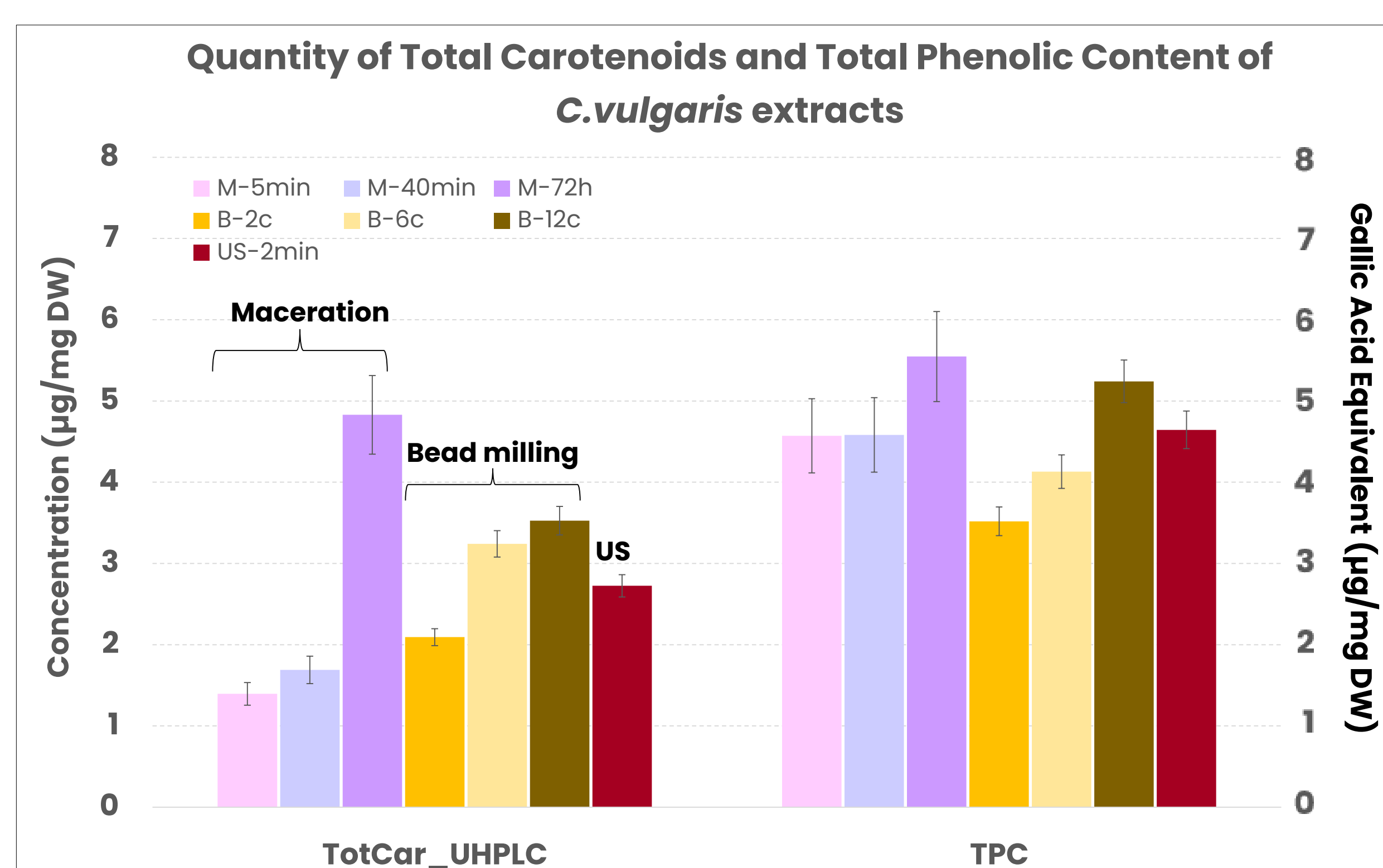
Pigment and Phenol Quantification

Material and methods

- UHPLC-DAD for pigment identification and quantification
- Total phenolic content (TPC) determined by Folin-Ciocalteu method (1927)



Chromatogram of *C.vulgaris* extracts obtained by UHPLC-DAD



Conclusions

Cell wall disruption

- Cell wall disruption was only achieved by coupling pure ethanol with bead milling
- 6 cycles of 2 min are enough to disrupt CCALA 924 cell wall

Extraction efficiency

- UHPLC was used to determine TotCar instead of spectrophotometry and Lichtenthaler equations for accuracy (Lichtenthaler, 1987)
- 72h maceration is the most efficient process for carotenoids and phenols extraction
- 6 bead milling cycles are enough to reach the maximum TotCar and ORAC and DPPH responses
- Ultrasound-assisted extraction enables the recovery of significant amounts of phenols and carotenoids relative to the applied extraction time

Antioxidant activity

- Lower response of the extracts for the DPPH assay
- The extracts' response depends on the assay : for example, the US-assisted extract has the lowest response with the ORAC test but the second highest one with DPPH
- Extracts with the highest carotenoid and phenolic content exhibit the most significant activity