Linking Bioaccessibility and Carotenoids to Antioxidant Activity in Diatoms

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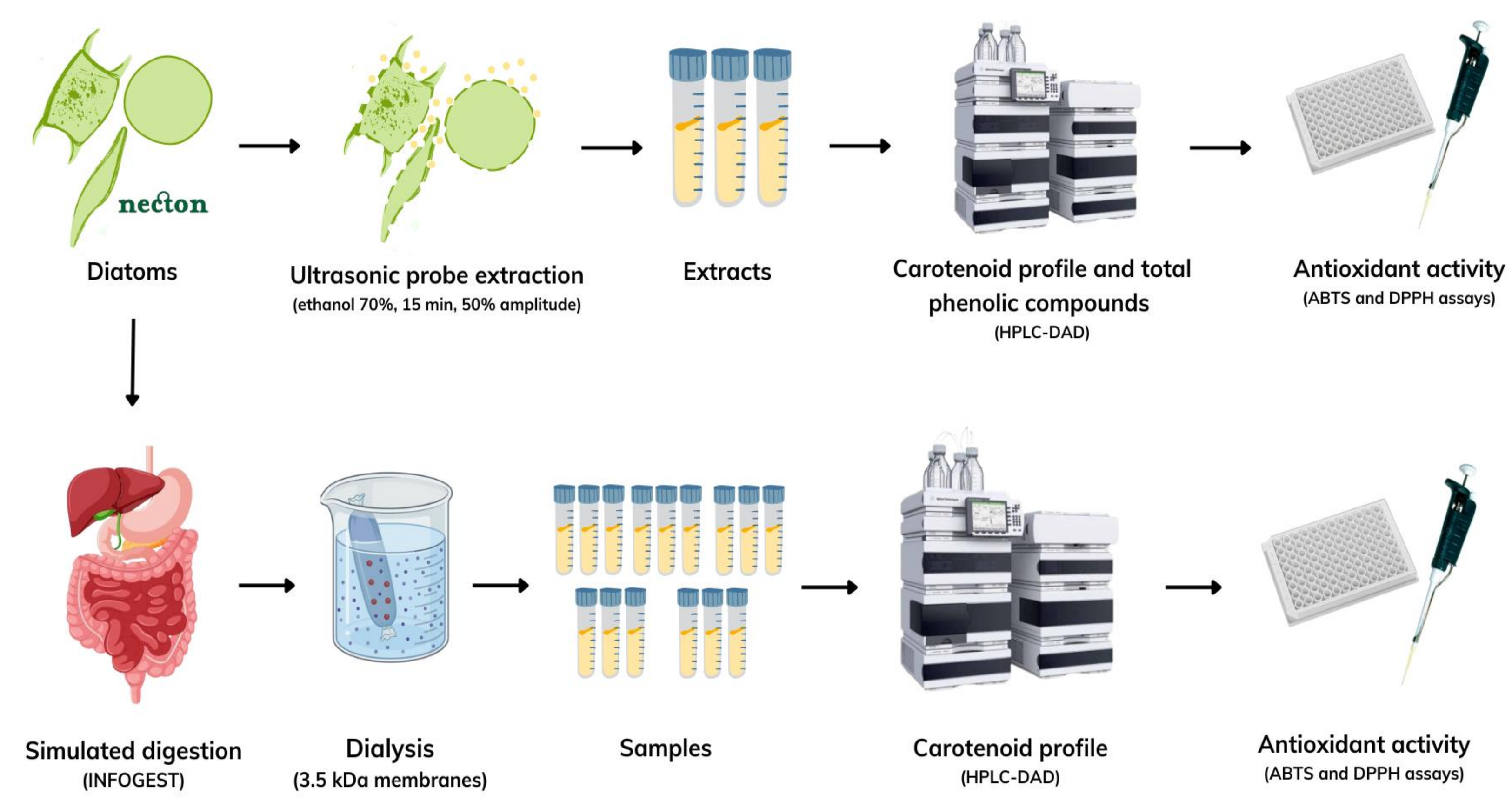
Introduction

Diatoms are photosynthetic microalgae with a siliceous cell wall and remarkable adaptability, making them a sustainable and renewable resource for high-value bioactive compounds (1,2). Their rapid growth, efficient carbon fixation, and minimal land and freshwater requirements (1,2) position them as attractive candidates for the development of eco-friendly products in the nutraceutical, pharmaceutical, and cosmetic industries. Among their most relevant metabolites are carotenoids and phenolic compounds, which exhibit strong antioxidant properties (3,4,5). This study explores the carotenoid profile of *Chaetoceros calcitrans*, *Nannofrustulum shiloi*, and *Cylindrotheca fusiformis*, and evaluates their antioxidant capacity using ABTS and DPPH assays. Furthermore, the natural bioaccessibility of carotenoids was assessed using the INFOGEST *in vitro* digestion model, highlighting the relevance of these compounds under physiological conditions and their potential for sustainable functional applications.

Objectives

- Characterise the total carotenoids and their profile and evaluate the antioxidant activity of the three diatom species;
- Assess the natural bioaccessibility of carotenoids, as well as the antioxidant activity of the diatoms in the post-dialysis fractions, to evaluate the role of the silica frustule as a barrier.

Methodology



Results and Main Conclusions

Table 1. Carotenoid profiles of *Nannofrustulum shiloi*, *Chaetoceros calcitrans*, and *Cylindrotheca fusiformis*. Carotenoid concentrations, total carotenoids and total chlorophylls are expressed as mg per g of biomass. Data represent mean values ± standard deviation (N=3).

	Concentration (mg/g biomass)		
Carotenoid	***N. shiloi***	***C. calcitrans***	***C. fusiformis***
Beta-cryptoxanthin	0.15 ± 0.02 c	0.39 ± 0.10 a	0.58 ± 0.03 b
Astaxanthin	0.25 ± 0.03 c	0.59 ± 0.07 a	0.41 ± 0.04 b
Fucoxanthin	0.59 ± 0.05 c	2.92 ± 0.17 a	1.13 ± 0.09 b
Zeaxanthin	0.12 ± 0.03 b	0.07 ± 0.003 a	ND
Total carotenoids	1.5 ± 0.05 c	6.8 ± 0.25 a	2.7 ± 0.09 b
Total Chlorophylls	3.3 ± 0.03 c	14.2 ± 0.41 a	12.2 ± 0.18 b

ND - Non-detectable.

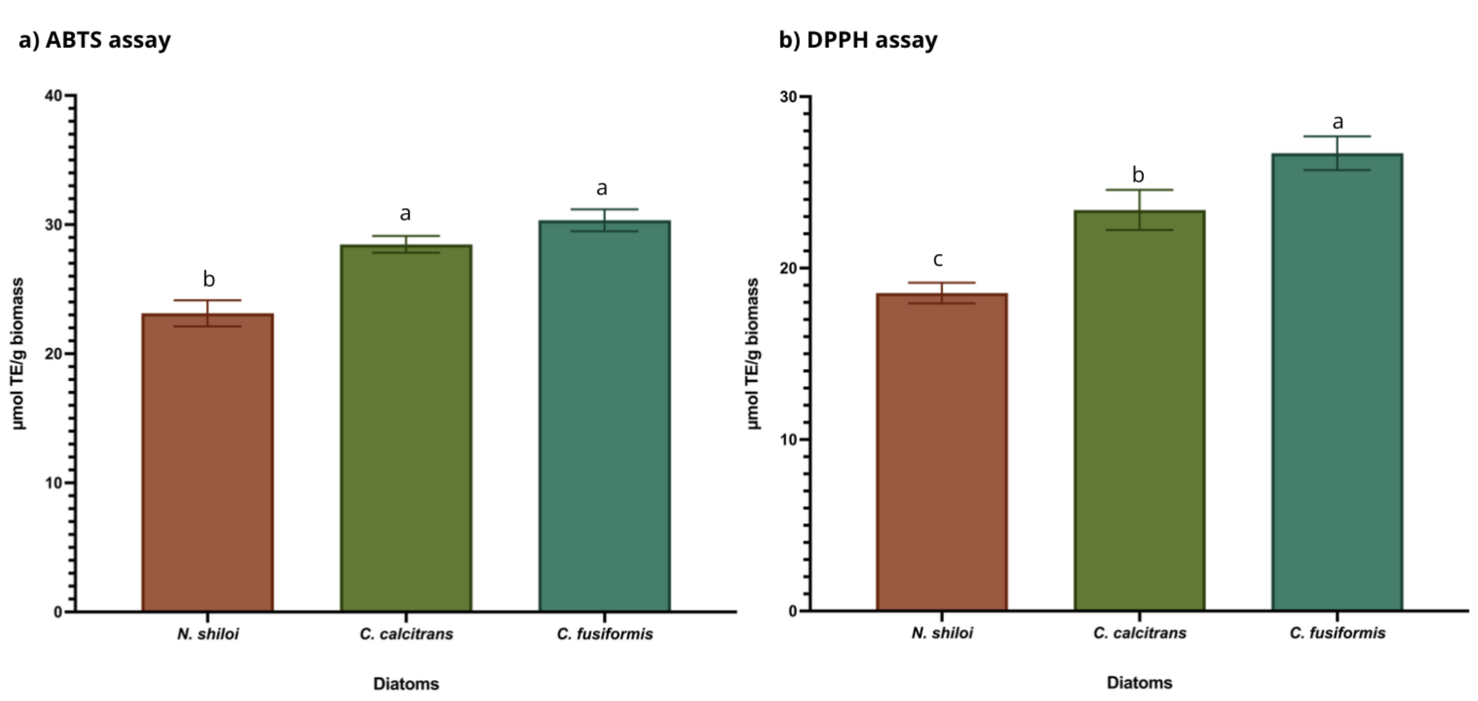


Figure 1. Antioxidant activity (µmol TE/g biomass) of ultrasound probe extracts from the three diatoms, measured by (a) ABTS, and (b) DPPH assays. Data are presented as mean values ± standard deviation (SD) from triplicate measurements.

- **Fucoxanthin** is the **most abundant carotenoid** across all species, with the highest concentration observed in ***C. calcitrans;***
- The diatoms ***C. calcitrans*** and ***C. fusiformis*** exhibit **higher antioxidant activity** in both assays (ABTS and DPPH) compared to ***N. shiloi***.

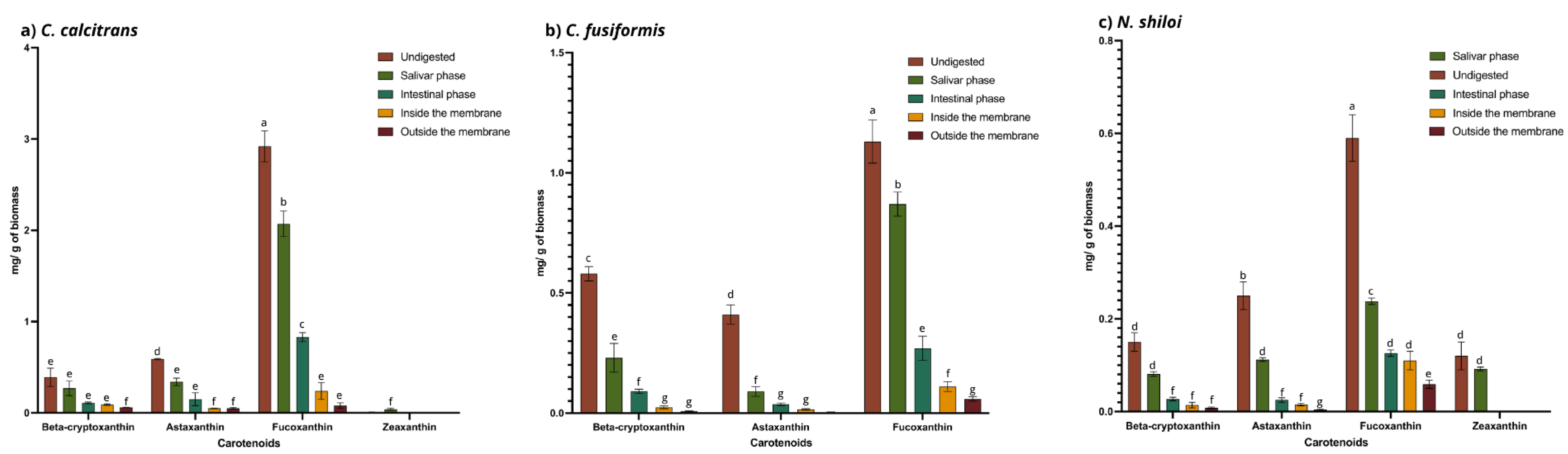


Figure 2. Carotenoids profile of a) *C. calcitrans*, b) *C. fusiformis*, and c) *N. shiloi* at the Undigested, salivar, intestinal, inside and outside of the dialysis membrane fractions with their respective concentrations (Mean ± standard deviation (SD)) in mg/g de biomass.

$$\text{Bioaccessibility index} = \frac{\text{Bioactive content in the intestinal phase}}{\text{Total bioactive content in the undigested sample}} \times 100$$

Carotenoids	*N. shiloi*	*C. calcitrans*	*C. fusiformis*
β-cryptoxanthin	5.21 %	18.21 %	15.65 %
Astaxanthin	10.07 %	15.42 %	9.07 %
Fucoxanthin	21.34 %	28.42 %	24.31 %
Zeaxanthin	-	-	-

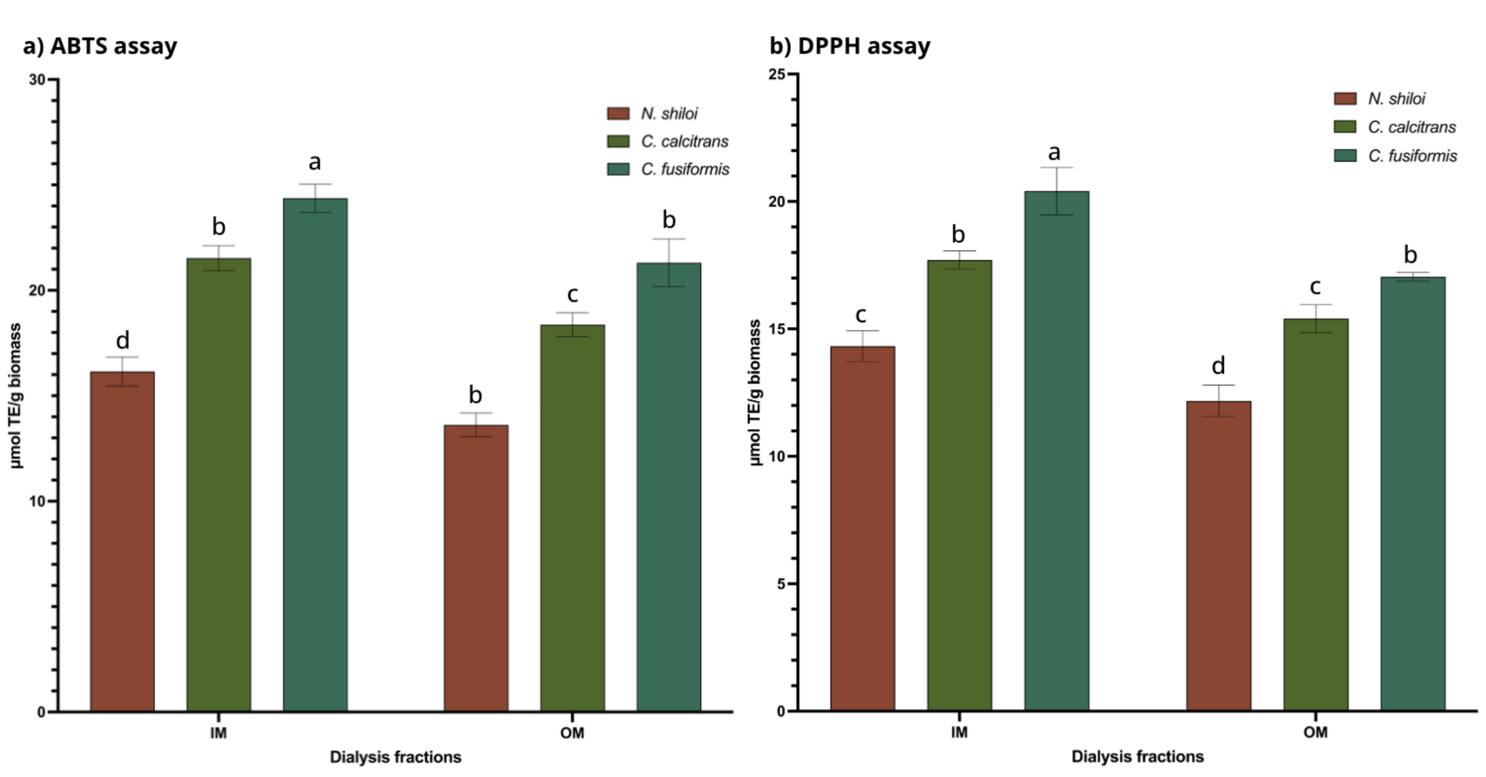


Figure 3. Antioxidant activity (µmol TE/g biomass) of samples from **inside of membrane (IM)** and **outside of membrane (OM)** for each digested diatom, measured by (a) ABTS, and (b) DPPH assays. Data are presented as mean values ± standard deviation (N=3).

- Both ***C. fusiformis*** and ***C. calcitrans*** exhibited **higher antioxidant activity** than ***N. shiloi*** in both ABTS and DPPH assays;
- **Antioxidant activity** was significantly **lower** in **OM** fractions compared to **IM** fractions across all species.

CONCLUSIONS

- The high antioxidant activity and good levels of carotenoids of the diatoms, highlight their potential for nutraceutical applications;
- The frustule acts as a physical barrier to biological potential: antioxidant activity and carotenoid concentration was lower;
- The bioaccessibility of carotenoids was below 50%, supporting the need for protection strategies to further enhance their health-promoting effects.

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References

1. Min, K.H., Kim, D.H., Youn, S., Pack, S.P., 2024. Biomimetic Diatom Biosilica and Its Potential for Biomedical Applications and Prospects: A Review. Int J Mol Sci 25, 2023. https://doi.org/10.3390/ijms25042023
2. Hopkinson, B.M., Dupont, C.L., Allen, A.E., Morel, F.M.M., 2011. Efficiency of the CO 2 -concentrating mechanism of diatoms. Proceedings of the National Academy of Sciences 108, 3830–3837. https://doi.org/10.1073/pnas.1018062108
3. Sardo, A., Orefice, I., Balzano, S., Barra, L., Romano, G., 2021. Mini-Review: Potential of Diatom-Derived Silica for Biomedical Applications. Applied Sciences 11, 4533. https://doi.org/10.3390/app11104533
4. Fu, W., Wichuk, K., Brynjólfsson, S., 2015. Developing diatoms for value-added products: challenges and opportunities. N Biotechnol 32, 547–551. https://doi.org/10.1016/j.nbt.2015.03.016
5. Maoka, T., 2020. Carotenoids as natural functional pigments. J Nat Med 74, 1–16. https://doi.org/10.1007/s11418-019-01364-x

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