Journal of Applied Phycology (2025) 37:2845–2858
https://doi.org/10.1007/s10811-025-03558-1

RESEARCH

Comparative analyses and characterization of eight seaweed species from the southeastern Bay of Biscay for potential biotechnological applications

Marina Salido[1,5] · Manu Soto[2,5] · Jone Bilbao[1,5] · Endika Quintano[1,5] · Néstor Etxebarria[3,5] · Inés Arana[4,5] · Maite Orruño[4,5] · Arkaitz Almaraz[4] · Saioa Gómez-Zorita[6,7,8] · María P. Portillo[6,7,8] · Sergio Seoane[1,5]

Received: 7 February 2025 / Revised: 16 May 2025 / Accepted: 18 May 2025 / Published online: 10 June 2025
© The Author(s) 2025

Abstract

The increasing interest in bioactive compounds from seaweed is demanding the characterization of local species, in order to know their suitability for different biotechnological or food applications. The present work aimed to characterize eight macroalgal species: *Codium decorticatum*, *Ulva rigida*, *Bifurcaria bifurcata, Halopteris scoparia, Chondracanthus acicularis*, *Gelidium corneum, Halopithys incurva* and *Porphyra linearis.* The species were collected in May, June and September 2022 and February 2023 in the Southeastern Bay of Biscay and microbiological and biochemical analyses were made, including pigments, mineral, metals, and protein and ash contents characterization. Concerning microorganisms, results showed that pathogenic or indicator microorganisms were only detected at low densities. Furthermore, the washing or freezing of macroalgae showed an almost total reduction of the studied microorganisms in both cases. Regarding pigments, *H. scoparia* stands out for the chlorophyll and fucoxanthin content and *P. linearis* for its β-carotene, lutein and phycobilins content. For minerals, maximum values were found in *B. bifurcata* and *H. incurva*. Additionally, according to European regulations, the recommended limits for both Hg and Cd metals were not exceeded. Finally, regarding proteins, the highest values were found mainly in the red macroalgae studied, which highlighted the use of seaweed as a natural protein source. These findings provide data about the characteristics of common seaweed regarding some bioactive compounds and their potential biotechnological applications.

Keywords Chlorophyta · Phaeophyta · Rhodophyta · Pigment · Metal · Protein · Microorganism

Introduction

Seaweeds (marine macroalgae) contain bioactive compounds that have demonstrated therapeutic properties for health and disease management (e.g. anticancer, antiobesity or antiviral), as well as certain nutrients that give them some nutritional benefits (Ioannou and Roussis 2009; Andrade et al. 2013). These bioactive metabolites with great potential have gained interest in different sectors of the industry, such as food, cosmetics, textile, agriculture and pharmaceutical (Shahnaz and Shameel 2009; Wells et al. 2017). Those compounds include proteins, amino acids, polysaccharides, and fatty acids as well as other secondary bioactive components such as phenolic compounds, vitamins, and pigments (Alisha et al 2019; Cherry et al. 2019; Siddique et al. 2022).

Seaweeds are an important source of proteins, which, together with peptides, show interesting bioactivities, with multiple applications as food and pharmaceutical compounds (Thiviya et al. 2022). The main protein types can be identified as enzymes, glycoproteins, lectins or phycobiliproteins (Echave et al. 2022). Furthermore, they also contain considerable quantities of pigments, which, besides exhibiting biological properties with potential benefits for human health, such as antioxidant, anticancer, anti-inflammatory, or biomedical applications, among others (Freitas et al. 2022), are a promising alternative in the pharmaceutical, agriculture, and textile sectors (Alisha et al. 2019) and as an artificial food coloring (Cikos et al. 2022).

In aquatic systems, seaweeds provide habitat structure and food for biological communities and among epiphytic microbes of seaweed, bacteria are very abundant.

Extended author information available on the last page of the article

Seaweed-associated bacterial communities not only vary among species, but also display temporal and spatial variations (Løvdal et al. 2021; Kalvaitienė et al. 2023) and bacterial communities could include pathogens such as *Vibrio* strains (Egan et al. 2013). The density of microorganisms on the fresh edible seaweed may serve as an indicator for the food quality and safety of the edible seaweed. The shelf life and sensorial quality of the seaweed could be affected negatively by high bacterial counts. Moreover, for some pathogens, consumption of relatively small amounts may be sufficient to cause severe health problems in humans. Thus, it is important to cover all basic information concerning essential characterization, not only to improve the multiple benefits, but also to know the possible weaknesses that could be present concerning seaweed (Mendes et al. 2022). One of these weaknesses is related to the lack of protocols regulating the use of those marine organisms. It is known that seaweeds accumulate contaminants from surrounding waters. Consequently, for the correct evaluation of seaweeds, it is necessary to know the possible risks concerning the presence and possible high concentrations of microorganisms, metals or iodine (Spillias et al. 2022), avoiding the generalization for the different species, which must be analyzed individually (Timoner et al. 2020).

The Basque Coast (NE Iberian Peninsula) is a habitat of approximately 372 macroalgal taxa (Gorostiaga et al. 2004). However, in this area, most of the macroalgal studies have been focused on the ecophysiology of species (Quintano et al. 2013, 2019), the effects of climate change or pollution on communities (Díez et al. 2012; Borja et al. 2013; Muguerza et al. 2017; Arriaga et al. 2024), or taxonomy (Secilla et al. 1997). In fact, although composition plays a significant role in determining the value of seaweed species from a nutritional and pharmaceutical point of view, it has not received enough research attention yet. This lack of detailed information delays the development as well as the potential utilization of these species, especially for gastronomic uses. In this context, this work aimed to analyze and characterize in detail eight local species from the southeastern Bay of Biscay, specifically the chemical (mineral and metals), biochemical (pigments, proteins and ash composition) and microbiological analyses (for human consumption). The goal is to provide a complete and comprehensive profile of these species, given special attention to the profitability of the compounds analyzed and their possible industrial approaches.

Material and methods

Study area, sampling collection and procedure

The studied area of the Southeastern Bay of Biscay (Cantabrian Sea), located in the northeast of the Iberian Peninsula (Fig. 1), is an open coast due to its exposure to the strong waves mainly coming from the northwest, and west-northwest (Díez et al. 2003; Galparsoro et al. 2010) that are responsible for the erosion of the coast. It is dominated by rocky substrata (Chust et al. 2011), suitable for macroalgal colonization, and it becomes sandy areas in deeper waters. Biogeographically, the area belongs to the warm sub-regions of the North-east Atlantic Ocean (Van den Hoek and Breeman 1990). The temperature used to be between 12 °C in winter and 22 °C in summer (Borja et al. 2000), although it has increased at a rate of 0.15 and 0.25 °C per decade, mainly at the surface (Chust et al. 2022; Laurenns-Balparda and Seoane 2024). The values of salinity are between 34 and 35.6 in the latest years (PdE 2024).

Based on previous studies on the distribution and diversity of macrophytes along the Basque coast (Gorostiaga et al. 2004), eight common species were selected because they are easily found in the area, considering their abundance, seasonality and place in the bathymetric range, being possible candidates in the zone if future uses would be considered. Furthermore, these species could show significant potential uses due to their biological properties, previously reported. The selected macroalgae were two species of class Ulvophyceae (Phylum Chlorophyta): *Codium decorticatum* (Woodward) M. Howe and *Ulva rigida* C. Agardh; two species of class Phaeophyceae (Phylum Heterokontophyta): *Bifurcaria bifurcata* R.Ross and *Halopteris scoparia* (Linnaeus) Sauvageau; and four species of phylum Rhodophyta: *Chondracanthus acicularis* (Roth) Fredericq, *Gelidium corneum* (Hudson) J.V. Lamouroux, *Halopithys incurva* (Hudson) Batters and *Porphyra linearis* Greville.

Samples were collected by hand in May, June and September 2022 and February 2023. For each species, approximately 60 g were collected in the lower intertidal zone of Lemoiz,

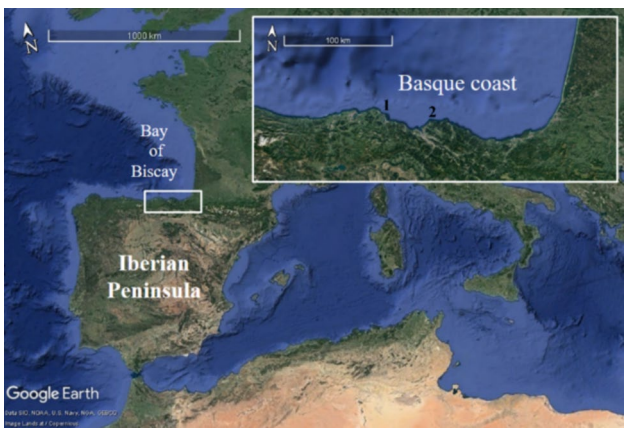


Fig. 1 Map of the study area and sampling points. **1**: Laredo, Cantabria (43º24′35.28'' N, 3º24′58.07'' W). **2**: Lemoiz, Basque Country (43º25259.993 N, 2º5222.1 W). Images obtained from Google Earth Pro

Basque Country (43°25′59.99″ N, 2°52′21″ W), except *C. decorticatum* and *G. corneum*, which were collected in Laredo, Cantabria (43°24′35.28"N, 3°24′58.07″ W) (Fig. 1) in the subtidal zone. The macroalgal collection involved selecting separate thalli along the sampling point to ensure a representative sampling of each beach and species. Samples were transported (30 min approximately) to the laboratory inside labelled plastic bags and kept in cooler boxes for macroalgal preservation and to prevent potential alterations that could affect the results. Once in the laboratory and before analyses, samples were washed with freshwater to remove epiphytes and other impurities. Then, samples were separated and taxonomically identified, confirming and supporting their identification and scientific nomenclature by experts and the databases AlgaeTraits (AlgaeTraits 2024) and AlgaeBase (Guiry and Guiry 2024). For each species, three replicates were randomly selected (from the 60 g of collected biomass) for the pigment analyses, and one replicate for the microorganism analyses. The remaining biomass was freeze-dried and homogenized for metal analyses (one replicate) and protein and ash analyses (two replicates each). The freeze-dried process was conducted in the Freezone plus 12 L, which was conducted from −40 to 21 °C for 50 h, and maintained until the total freeze-dry state in 3 to 5 days.

Pigment extraction

Lipophilic pigments (chlorophylls and carotenoids) were extracted and analyzed by High-Performance Liquid Chromatography (HPLC) following Zapata et al. (2000), with modifications and the equipment detailed in Seoane et al. (2009). For each species, three randomly selected replicates of around 0.015 g each were analyzed and the quantification of the pigments was made based on commercial standards. For the pigments that were not calibrated with commercial standards, molar extinction coefficients were used (Jeffrey et al. 1997).

For the extraction of hydrophilic pigments (PBP), three randomly selected replicates (0.2–0.5 g each) from each species were cryogenized and, under low light conditions, ground with a Retsch Mixer Mill MM 400. The resulting powders were added in a Falcon tube with 5 mL of 0.1 M potassium phosphate buffer pH 7. The samples were centrifuged at 4 °C, 5000 rpm for 15 min. The supernatant was used for phycobiliprotein determinations measuring the absorbance at 455, 564, 592, 618 and 645 nm to quantify phycoerythrin (R-PE) and phycocyanin (R-PC) content. The two following equations of Beer and Eshel (1985) were used:

$$\text{R-PE} = \left[\left(A_{564} - A_{592}\right) - \left(A_{455} - A_{592}\right)x0.2\right]x0.12 = \text{mg mL}^{-1}$$

$$\text{R-PC} = \left[\left(A_{618} - A_{645}\right) - \left(A_{592} - A_{645}\right)x0.51\right]x0.15 = \text{mg mL}^{-1}$$

Concentrations of all pigments were transformed to mg g^{-1} dry weight (DW) with the relation DW WW^{-1} (wet weight), which is the result of the drying process of macroalgae for 24 h at 60 °C in a drying oven.

Microbiological parameters

Total mesophilic aerobes, halophiles, *Staphylococcus aureus*, sulfite-reducing clostridia, *Clostridium perfringens*, *Vibrio cholerae*, enterobacteria, coliforms and *Escherichia coli* microbiological parameters were determined for each species. Analyses were carried out directly (< 24 h from collection) to minimize the risk of microbial alterations. One replicate of 25 g of each species was transferred into sterile Stomacher bags and homogenized with both 225 mL of alkaline-saline *Vibrio* enrichment broth (alkaline-peptone water [APSW], Merck) for the detection of enteropathogenic *Vibrio cholerae*, or with 225 mL of peptone saline water (PSW, Merck) for all other quantifications. For *V. cholerae* detection, fresh macroalgal samples were incubated for 6 h at 41.5 °C. Detection of *Vibrio* in enrichment broth was carried out following ISO 21872–1:2017 (Annex 1). The aerobic mesophiles microorganisms were estimated according to ISO 4833–1:2013 (Annex 1) and aerobic halophilic mesophiles were enumerated by supplementing the culture medium with 2% NaCl (w/v). Enterobacteria, coliforms, *Escherichia coli, Staphylococcus aureus* and *Clostridium perfringens* were enumerated following ISO 21528–2:2017, ISO 4832:2006 (reviewed and confirmed in 2021), ISO 16649–2:2011, ISO 6888–1:2022 and ISO 15213–2:2023, respectively (Annex 1). Finally, sulphite-reducing clostridia were determined by using sulfite polymixin sulfadiazyne (SPS) agar (Condalab) incubated for 24 h at 37 °C in anaerobiosis.

In addition, *B. bifurcata, H. scoparia* and *H. incurva* were selected to analyze how common processing treatments, such as freezing and washing, affect microbial counts. For this purpose, microbiological quantifications for aerobic mesophiles, aerobic halophilic mesophiles and enterobacteria, previously detailed, were repeated after two treatments: fresh washed (W) and frozen unwashed (FR), and the results were compared with fresh unwashed control samples (F). The washes were performed with a gentle tap water wash (1 min), and the frozen samples were stored in a conventional freezer (−20 °C) for approximately 1 month.

Mineral and metal analysis

Elemental analysis of Al, As, Ca, Cd, Co, Cr, Cu, Fe, Hg, K, Mg, Mn, Mo, Na, Ni, Pb, Sb, Si, Sn and Zn was carried out by ICP-MS (NexIon200, Perkin Elmer) for each species. One replicate (0.1–0.2 g) of freeze-dried biomass for each species were digested in a microwave oven as described

by Bartolomé et al. (2010). Then, the samples were sipped with a peristaltic pump to the nebulization system (Meinhard nebulizer) and simultaneously, through a parallel line a continuous flow of internal standard solution (Ge, Sc and Y), was mixed and measured to overcome measurement drifts. The calibration set was measured three times and the six blanks were prepared and measured to estimate detection limits. However, the samples were measured only once.

Protein composition

The protein analyses of the macroalgae were analyzed according to the Association of Official Analytical Chemists (AOAC) Official Methods (Cunniff 1995).

Protein content was measured by a modification of the Kjeldahl method using a conversion factor for nitrogen to protein of 4.59 (Kjeldahl 1883; Cruz 2013). For each species, two replicates (approximately 0.2 g each) of freeze-dried biomass were digested by sulphuric acid in the presence of a catalyst for 3 h at 400 °C. Then, ammonia was distilled in a vessel containing boric acid and an acidimetric indicator. Finally, the titration was carried out with 0.1 N standardized hydrochloric acid.

Due to the sampling size limitation, *C. acicularis* was not measured.

Ash composition

The ash composition of the macroalgae were analyzed according to the Association of Official Analytical Chemists (AOAC) Official Methods (Cunniff 1995).

For ash determination, two replicates (approximately 0.4 g each) of freeze-dried biomass were incorporated in a conventional muffle for each species. The temperature of the muffle was slowly increased until reaching 650 °C, a temperature that was maintained for 6 h to ensure the constant weight was gained. Ash content was finally weighed.

Due to the sampling size limitation, *C. acicularis* was not measured, and for *U. rigida* and *G. corneum,* only one replicate was analyzed.

Data analysis

The differences in the pigment profiles of the studied species and the effect of the sample-treatment on the microorganism content of the macroalgae were statistically tested. Statistical analyses were done using PAST 3.26 (Paleontological Statistics software package for education and data analysis). After validating the normality and homogeneity of variances, differences in the pigment profiles of the studied species were analyzed with a one-way PERMANOVA, with an a priori chosen significance level of $\alpha = 0.05$. Furthermore, in order to know the specific differences among pigments between all studied species, a non-parametric Kruskal–Wallis test was carried out. For the pigments Chl *c*, Neox and Fuco, the Mann–Whitney test was performed, instead of Kruskal–Wallis, since they were present just in two species (a two-sample comparison). On the other hand, the effect of sample-treatments (washed, fresh or frozen; W, FR and F) on microorganisms from *B. bifurcata, H. scoparia* and *H. incurva* was evaluated by comparing W-FR, W–F and FR-F with a Wilcoxon test ($\alpha = 0.01$), analyzing all the data from the studied species together. Microorganisms, mineral, metals and protein and ash composition quantification were not subjected to statistical analysis as they were done for basic characterization purposes.

Results

Pigments profile

The pigment analyses showed the usual pigment profile of each taxonomic group (Table 1), showing significant differences in the overall pigment-content ($p < 0.05$) among the studied species (Annex 2). Additionally, these differences were also observed for some of the pigments individually. Regarding chlorophylls, both Chl *a* and *b* showed significant differences ($p < 0.05$) among the studied species (Annex 3) For Chl *a,* the highest values were found in *C. decorticatum* and *H. scoparia* (Phaeophyceae, Heterokontophyta) with 5.09 ± 0.65 and 5.41 ± 1.03 mg g^{-1} DW, respectively. In contrast, *C. acicularis* showed the lowest values (0.17 ± 0.10 mg g^{-1} DW). Concerning Chl *c*, no significant differences ($p < 0.05$) were observed among the studied species (Annex 3). Moreover, in line with the literature, they were just present in the green and brown macroalgae, respectively.

Concerning carotenes, α- and β-carotene were found in the present study, the latter showing significant differences ($p < 0.05$) among studied species (Annex 3). All the analyzed species shared β-carotene, except *C. decorticatum* (Table 1), with *P. linearis* the one which showed the highest values (0.34 ± 0.24 mg g^{-1} DW) and *C. acicularis* the lowest (0.01 ± 0.01 mg g^{-1} DW). Regarding xanthophylls, lutein and violaxanthin showed significant differences ($p < 0.05$) among the species that contained these pigments (Annex 3) (Table 1). Lutein was the dominant xanthophyll found in *P. linearis* and *H. incurva* (Rhodophyta), being also the ones which showed maximum values of this pigment, 0.85 ± 0.13 and 0.32 ± 0.12 mg g^{-1} DW, respectively. For fucoxanthin and violaxanthin, the highest results were observed in *H. scoparia* (Phaeophyceae, Heterokontophyta) (2.22 ± 0.45 and 0.21 ± 0.03 mg g^{-1} DW, respectively).

Regarding phycobiliproteins, phycocyanin (PC) and phycoerythrin (PE) were only found in the red macroalgae, being the values of PE higher than PC (Table 1), ranging PE

Table 1 Pigment profile for the selected macroalgal species. Concentrations are expressed in mg g^{-1} DW of mean ± standard deviation (n = 3). Significant differences between treatments are shown with different letters. Chl: Chlorophyll. Car: carotene. Anthera: Antheraxanthin. Lut: Lutein. Neox: Neoxanthin. Sipho: Siphonoxanthin. Diadino: Diadinoxanthin. Sipho-d: Siphonoxanthin dodecanoate Viola: Violaxanthin. Zea: Zeaxanthin. Fuco: Fucoxanthin. PE: phycoerytrhin. PC: phycocyanin. ND: non-detected

			Chlorophyta		Phaeophyceae		Rhodophyta			
			U. rigida	*C. decorticatum*	*B. bifurcata*	*H. scoparia*	*G. corneum*	*P. linearis*	*C. acicularis*	*H. incurva*
CHLS	Chl *a*		1.80 ± 0.69 ^{ab}	5.09 ± 0.65 ^{a}	1.35 ± 0.35 ^{ab}	5.41 ± 1.03 ^{a}	1.63 ± 0.40 ^{ab}	4.19 ± 0.23 ^{ab}	0.17 ± 0.10 ^{b}	1.18 ± 0.68 ^{ab}
	Chl *b*		1.11 ± 0.42 ^{ab}	3.41 ± 0.43 ^{a}	ND	ND	ND	ND	0.00 ± 0.01 ^{b}	0.13 ± 0.01 ^{ab}
	Chl *c2*		ND	ND	0.07 ± 0.01 ^{a}	0.27 ± 0.04 ^{a}	ND	ND	ND	ND
	Chl *c1*		ND	ND	0.02 ± 0.00 ^{a}	0.17 ± 0.03 ^{a}	ND	ND	ND	ND
CAROTENOIDS	Car	α – Car	ND	0.33 ± 0.04 ^{a}	ND	ND	0.13 ± 0.03 ^{a}	ND	ND	0.16 ± 0.06 ^{a}
		β—Car	0.03 ± 0.02 ^{ab}	ND	0.06 ± 0.01 ^{ab}	0.18 ± 0.04 ^{a}	0.03 ± 0.00 ^{ab}	0.34 ± 0.24 ^{ab}	0.01 ± 0.01 ^{b}	0.08 ± 0.03 ^{ab}
	Xanthophylls	Anthera	ND	ND	ND	0.01 ± 0.00	ND	ND	ND	ND
		Lut	0.16 ± 0.07 ^{ab}	ND	ND	ND	0.17 ± 0.04 ^{ab}	0.85 ± 0.13 ^{a}	0.01 ± 0.01 ^{b}	0.32 ± 0.12 ^{ab}
		Neox	0.00 ± 0.00 ^{a}	0.23 ± 0.03 ^{a}	ND	ND	ND	ND	ND	ND
		Sipho	ND	0.53 ± 0.07	ND	ND	ND	ND	ND	ND
		Diadino	ND	ND	ND	0.02 ± 0.00	ND	ND	ND	ND
		Sipho-d	ND	0.43 ± 0.05	ND	ND	ND	ND	ND	ND
		Viola	0.02 ± 0.01 ^{a}	0.03 ± 0.00 ^{ab}	0.04 ± 0.02 ^{ab}	0.21 ± 0.03 ^{b}	ND	ND	ND	ND
		Zea	ND	ND	ND	0.03 ± 0.02 ^{a}	ND	0.14 ± 0.12 ^{a}	0.00 ± 0.00 ^{a}	ND
		Fuco	ND	ND	0.40 ± 0.04 ^{a}	2.22 ± 0.45 ^{a}	ND	ND	ND	ND
PBP	PE						3.15 ± 3.03 ^{ab}	10.89 ± 0.71 ^{a}	0.10 ± 0.07 ^{b}	0.78 ± 0.07 ^{ab}
	PC						0.20 ± 0.16 ^{ab}	3.32 ± 0.30 ^{a}	0.22 ± 0.13 ^{ab}	0.06 ± 0.04 ^{b}

concentrations from 0.10 ± 0.07 mg g^{-1} DW in *C. acicularis* to 10.89 ± 0.7 mg g^{-1} DW in *P. linearis*. Furthermore, Both PC and PE showed significant differences ($p < 0.05$) among red seaweed studied (Annex 3), being the highest abundances of both pigments detected in *P. linearis*, with concentrations of 3.32 ± 0.3 and 10.89 ± 0.7 mg g^{-1} DW, respectively.

Microbiological parameters

Direct analysis (< 24 h from collection) of microbiological parameters revealed considerable differences among the different macroalgae (Table 2). The highest number of mesophilic and halophilic bacteria was detected in *P. linearis* (1.2×10^5 and 1×10^5 CFU g^{-1}, respectively), followed by *C. acicularis* and *U. rigida* (above 10^4 CFU g^{-1}). In contrast, the lowest counts corresponded to *B. bifurcata* (below 10^3 CFU g^{-1}). Regarding pathogenic microorganisms or indicators of contamination, they were detected but always at low densities (Table 2). Enterobacteria was enumerated in most macroalgae, with the highest counts in *U. rigida* (1.3×10^3 CFU g^{-1}), but coliforms and *E. coli* only appeared, close to their detection limit, in *P. linearis* and *U. rigida* and *C. acicularis*, respectively. In the case of sulfite reducing clostridia, they were found in low numbers in *P. linearis*, *C. acicularis* and *H. incurva*, whereas *C. perfringens* was present in *U. rigida* and *C. acicularis.* Among other potential pathogens, *S. aureus* was only observed in *C. acicularis* (15 CFU g^{-1}) and *H. incurva* (10 CFU g^{-1}) and, in no case, *V. cholerae* was detected.

On the other hand, when evaluating how algae washing (1 min under running water) and freezing (−20 °C) affect microbial determinations, significant differences ($p < 0.01$) were shown among F-W and F-FR treatments (Annex 4), while FR-W shown there are not significant differences among treatments (Fig. 2). A generalized reduction of both mesophilic and halophilic microorganisms was observed and enterobacteria were no longer detected in both treatments (Fig. 2). In addition, no other indicator microorganisms or potential pathogens were recorded, except for 5 CFU g^{-1} of sulfite-reducing clostridia in washed samples of *H. incurva* and *H. scoparia*.

Mineral and metal accumulation

Despite values being variable among the species for all the elements, the average concentrations determined in the species of this study were in increasing order as follows: Sn < Sb < Cd < Hg < Mo < Co < Cr < Pb < Ni < Cu < As < Zn < Mn < Si < Al < Fe < Mg < Na < K < Ca. The lowest ranges were for Sn (0.04—0.09 µg g^{-1} DW), Sb (0.06—0.1 µg g^{-1} DW) and Hg (0.07—0.19 µg g^{-1} DW) and the highest for the macroelements Ca (1941—111,697 µg g^{-1} DW), K (134,086—65,059 µg g^{-1} DW) and Na (13,174—48,589 µg g^{-1} DW). *H. incurva*, followed by *B. bifurcata* were the species that accumulated the highest quantities, while *U. rigida* accumulated the least (Table 3). On the other hand, other interesting elements, from a nutritional point of view, are present in high quantities in some of the studied species. Fe, Mg reached up to 14,480 and 38,247 µg g^{-1} DW in *B. bifurcata* and *U. rigida*, respectively.

Protein content

The protein content of the macroalgae is presented in Table 4. The lowest protein content was found in green macroalgae *U. rigida* (2.95% DW) and brown macroalgae *B. bifurcata* (3.15% DW), whereas the highest was found in red macroalgae, ranging from 11.22% DW in *H. incurva* to 16.27% DW in *P. linearis.*

Table 2 Enumeration of microorganisms in fresh samples of the selected species. Values expressed in CFU g^{-1} (n = 1). ND: non-detected. (-): not analyzed. < 5: below of limit detection

	Chlorophyta		Phaeophyceae		Rhodophyta			
	U. rigida	*C. decorticatum*	*B. bifurcata*	*H. scoparia*	*G. corneum*	*P. linearis*	*C. acicularis*	*H. incurva*
Total mesophilic aerobes	1.8×10^4	-	9.8×10^2	2.7×10^3	-	1.2×10^5	4.1×10^4	2.9×10^3
Halophiles	1.7×10^4	-	6.6×10^2	2.6×10^3	-	1×10^5	5.1×10^4	5.3×10^3
S. aureus	< 5	-	< 5	< 5	-	< 5	15	10
Sulfite reducing clostridia	< 5	-	< 5	< 5	-	15	15	5
C. perfringens	5	-	< 5	< 5	-	< 5	5	< 5
V. choleae	ND	-	ND	ND	-	ND	ND	ND
Enterobacteria	1.3×10^3	-	20	90	-	40	300	40
Coliforms	< 5	-	< 5	< 5	-	5	< 5	< 5
E. coli	10	-	< 5	< 5	-	< 5	5	< 5

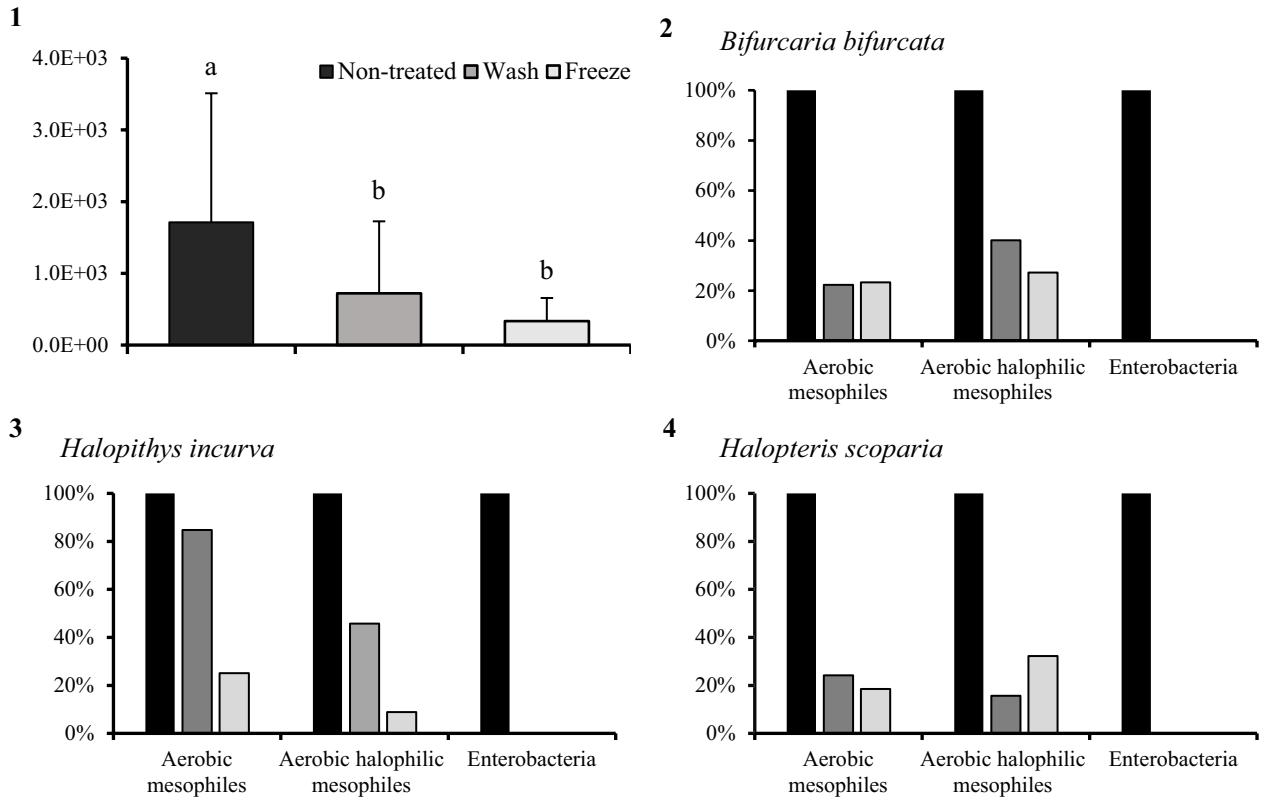


Fig. 2 Comparison of microorganism content after washing (▩) and freezing (□) with respect to non-treated fresh samples (■). **1**: Comparison taking into account the three selected algae and all the microorganisms species studied. Data are expressed as the mean ± standard deviation (CFU g^{-1}). Significant differences between treatments are shown with different letters. **2 to 4**: Specific comparison of aerobic mesophile, aerobic halophilic mesophiles and enterobacteria for *B. bifurcata, H. incurva* and *H. scoparia*, respectively. Data are expressed as the percentage (%)

Ash content

The ash content of the macroalgae is presented in Table 4. As far as ash content is concerned, the higher values were found in *U. rigida*, *C. decorticatum* and *H. scoparia*, 58.29, 47.16 and 56.33% DW, respectively, and the lowest values in *G. corneum* and *P. linearis*, 13.6 and 24.58% DW, respectively.

Discussion

The studied species were widely available for collection in the Basque Coast, although they appear to have specific seasons when they are most easily found. It is known that the biochemical and microbiological profile of seaweed is influenced not only by taxonomic groups, but also by biotic and abiotic factors such as life cycle, season, geographical location or depth. Therefore, although our working hypothesis in this study focused on differences in the basic microbiological, chemical, protein, ash, and pigment composition according to species, it is emphasized that the conditions (and sampling strategy) in which these species were collected should be considered when discussing present findings and comparing them with other studies.

Pigment profile

The differences among the pigment values of each analyzed species emphasize the importance of the characterization of the seaweed species as well as from different areas or periods of the year. In this context, among chlorophylls, *H. scoparia*, which in the results of the present study increased up to 5.41 ± 1.03 mg g^{-1} DW, is slightly lower when compared with data from Nunes et al. (2020) (8.15 mg g^{-1} DW). This discrepancy is due to the changes in sunlight hours and temperature, which drive shifts in photosynthetic activity, leading to adjusting pigment levels to optimize light absorption and protect against environmental changes (Cikos et al. 2022). In this case, algae increased chlorophyll levels production in the high irradiance months in which they were collected (Madeiran Archipelago in the 2017 spring).

Regarding carotenoids, something similar occurs with the β-carotene quantities found in the present study. For

Table 3 Mineral and metals analyzed for the selected species. Concentrations expressed in μg g^{-1} DW (n = 1). Detection limits were estimated from the average of six blank samples plus three times the standard deviation. (-): not analyzed

	LoD	Chlorophyta		Phaeophyceae		Rhodophyta			
		U. rigida	*C. decorticatum*	*B. bifurcata*	*H. scoparia*	*G. corneum*	*P. linearis*	*C. acicularis*	*H. incurva*
Al	0.33	98	-	1211	578	-	-	249	877
As	0.002	3.9	18.4	53	13	8.4	18.4	8.2	12
Ca		34,577	-	19,041	54,492	-	-	33,120	111,697
Cd	0.002	0.06	0.1	0.05	0.1	0.2	0.1	0.02	0.2
Co	0.005	0.2	-	1.2	0.9	-	-	0.6	1.4
Cr	0.02	0.4	0.5	1.8	1.4	3.8	0.1	0.4	1.3
Cu	0.04	4.5	1.9	4.7	5.4	6.0	4.6	4.2	5.2
Fe	0.7	147	-	14,481	1580	-	-	979	4312
Hg	0.05	0.1	0.2	0.05	0.07	0.2	0.2	-	0.06
K	0.3	20,103	-	64,642	24,411	-	-	13,086	65,059
Mg	0.1	38,247	-	6241	9842	-	-	9798	8148
Mn	0.01	11	-	74	102	-	-	50	165
Mo	0.002	0.08	-	0.4	0.4	-	-	0.13	0.9
Na	0.25	48,589	-	14,374	34,048	-	-	43,284	13,174
Ni	0.1	1.3	2.5	3.2	6.3	4.4	0.6	1.4	6
Pb	0.03	0.78	0.54	2.8	5.7	6.5	0.5	1.8	7
Sb	0.002	-	0.1	0.1	0.06	0.07	0.1	0.03	0.08
Si	0.25	-	-	228	390	-	-	88	486
Sn	0.004	0.09	0.06	0.04	0.05	0.04	0.07	-	0.05
Zn	0.025	16.4	6.6	28	61	104	32	26	51

Table 4 Protein and ash composition for the selected macroalgal species. Data are expressed in percentage (%) DW of mean ± standard deviation (n = 2). (-): not analyzed

	Chlorophyta		Phaeophyceae		Rhodophyta			
	U. rigida	*C. decorticatum*	*B. bifurcata*	*H. scoparia*	*G. corneum*	*P. linearis*	*C. acicularis*	*H. incurva*
Protein content	2.95 ± 0.64	6.75 ± 0.01	3.15 ± 0.45	7.21 ± 0.95	13.45 ± 0.98	16.27 ± 0.15	-	11.22 ± 1.36
Ash content	58.29	47.16 ± 1.64	24.58 ± 3.68	56.33 ± 1.42	13.6	16.63 ± 0.84	-	37.71 ± 0.85

P. linearis, the species with the highest β-carotene levels among those analyzed (0.34 ± 0.24 mg g^{-1} DW), conversely to the reported by Osório et al. (2020) and Schubert et al. (2006). For instance, Schubert et al. (2006) showed that *Porphyra perforata* showed 49.5% β-carotene of the total carotenoid content, whereas in the present study 25.5% β-carotene of the total carotenoid was obtained for *P. linearis*. These differences might be attributed to two main aspects: the differences in pigment profile, even among species of the same genus, or the nature of the pigment. Carotenoids not only serve as light-harvesting pigments, but also function as photoprotective pigments (Esteban et al. 2009) Their activation under high irradiance levels may explain the differences observed when comparing the environmental conditions of the Gulf of Mexico (Schubert et al. 2006) with present results.

Furthermore, among carotenoids, it is important to mention fucoxanthin, a pigment that has been deeply studied due to its multiple applications in the cosmetic, food nutraceutical, and pharmaceutical industries (Salido et al. 2024). In line with the literature, it was present in both Phaeophyceae studied species *H. scoparia* and *B. bifurcata*. These values also varied from those reported by other authors. For *H. scoparia*, Nunes et al. (2020) reported a six times lower value if compared with those observed in this study. In this case, these differences could be attributed not only to the nature of the pigments or environmental factors, but also to the extraction methods because they significantly influence the extraction efficiency, purity, and economic viability of the process, as reported by Leong et al. (2022).

Concerning phycobiliproteins, as expected, all the Rhodophyta species studied contained both PE and PC. However,

they showed marked differences between them, with *P. linearis* the one showing the highest values. As 470–660 nm range light harvesting pigments, PBP are more active and absorbs sunlight at low irradiances when the primary pigments are less efficient. In this sense, the highest PE and PC values showed in *P. linearis* (10.9 ± 0.7 and 3.3 ± 0.3 mg g^{-1} DW, respectively) could be explained by the fact this species was collected in February while the other three red algae were collected in summer seasons when the irradiance rise considerably in the zone of study. Moreover, considering comparisons with the results obtained in the literature, in the present research this species showed considerably high amounts if compared with other authors (Munier et al. 2013; Ji et al. 2023). Cotas et al. (2020) propose *Gracilaria gracilis*, with 7 mg g^{-1} DW PE, as a novel industrial source of PBP, which ranges between 50–146 € per mg, depending on the purity. In this sense, *P. linearis*, which showed higher values, could be proposed as a valuable source of PBP. However, although there are good yields for PBP from *P. linearis* and *G. gracilis*, other aspects could be taken into account, such as organisms that exhibit better yields, like cyanobacteria (Hemlata and Fatma 2009; Ji et al. 2023), or the instability of phycobiliproteins when exposed to adverse conditions (e.g. temperature or light) (Nowruzi et al. 2022), which is a critical aspect for industrial applicability. Therefore, further research focusing on the need for strategies to protect these pigments from degradation, the improved yield and extractions of phycobiliproteins from red macroalgae is needed (Cotas et al. 2020).

The study of seaweed pigments is essential and has been extensively researched due to their biological properties and their key role in photosynthesis. Macroalgae could be an excellent source for obtaining these pigments (Cikos et al. 2022), but harvesting them directly from nature is often neither feasible nor sustainable, and production costs remain high. For instance, in the case of chlorophyll extraction at an industrial scale, it is still more cost-effective to extract them from spinach or alfalfa (Chen and Roca 2018), as their production is significantly cheaper. For fucoxanthin, which is widely known for its antioxidant properties (Leong et al. 2022) it has already been commercialized, among others, by Algatech in Israel. However, the product they commercialize, FucoVital, which is associated with supporting liver health, uses the pigment extracted from microalgae, which is more efficient. For phycobiliproteins, although other organisms, like cyanobacteria, generally show a higher yield, *P. linearis* could serve as a source due to its high PE and PC content. In this context, regarding resource optimization, several studies point out the lack of further research that might help improve the production of pigments from macroalgae, focusing on the innovation of bioengineering techniques as well as the implementation of low-cost strategies for production (Leong et al. 2022).

Microbiological parameters

Most microorganisms in environmental samples are not detected when cultivation procedures are used, including pathogen microorganisms (Atlas and Bartha 1998). Moreover, methods of detection of pathogens are laborious and time-consuming, and it is difficult or costly to determine the presence of a specific pathogen. Detection of pathogens associated with fecal contamination is, therefore, a multifaceted challenge, which can be overcome by using fecal biological indicators to approximate many potential pathogens.

In the case of the food industry, Spain lacks specific regulations on microbiological analyses and maximum allowed limits for seaweed. The regulations of other countries do specify these requirements and, although there are differences among them, France stands out as the first European country to do a detailed evaluation of microbial limits in the consumption of macroalgae as food. Analyzing our data based on the requirements of the French Food Safety Agency (CEVA 2020), both coliforms and *S. aureus* are below the recommended limit, < 10 and $< 10^2$ CFU g^{-1} DW, respectively. However, some other limits have been exceeded, such as that of *C. perfringens* which was detected in *U. rigida* and *C. acicularis* (5 CFU g^{-1}) and whose maximum allowed limit is < 1 CFU g^{-1} DW. For aerobes mesophiles, whose limit is $< 10^5$ CFU g^{-1} DW, only *P. linearis* exceeded it in the fresh sample (1.2×10^5 CFU g^{-1}) (CEVA 2020).

Nevertheless, the data seem to indicate that pre-treatment, such as washing or freezing, could contribute to lowering this number under the recommended French limit. It is crucial to carry out standardized protocols and guidelines that establish maximum levels and treatments for microbial pathogens to ensure the safety of the product for consumers.

Mineral and metal content

Consuming seaweed may be linked to the transfer of high-risk metals to humans and, due to this different regulations establish some limits for this consumption. In Europe, the only requirement established by the European Union Recommendation (EU) 2018/464 of the Commission of 19 March 2018 recommends controlling the presence of arsenic, cadmium, lead, and mercury in seaweed, halophilic plants, and products based on seaweed. However, there is only a limit value for Cd and Hg. For Cd, this value has a maximum content of 3 µg g^{-1} dry weight (DW) in European Regulation No 915/2023 (European-Comission 25 April 2023). For Hg, the limit value has a maximum content of 0.01 mg kg^{-1} (European-Commission 19 March 2018). However, this regulation does not specify whether the content refers to fresh or dry weight of the macroalgae. Apart from this, France is the only European country that has a specific assessment of maximum levels of metals (inorganic arsenic, cadmium,

lead, tin and mercury) authorized in algae for human consumption (CEVA 2020).

Regarding the results of the present study and based on the European Regulations previously detailed, both Hg and Cd are below the recommended limits, 0.01 and 3 mg kg^{-1} (= μg g^{-1}), respectively, assuming these limits as DW. For the total As, highly variable values were shown, ranging from 3.8 to 53.3 μg g^{-1} DW. However, as previously commented,, the present As includes both organic and inorganic forms and knowing that each form has distinct effects on human health, they should be analyzed separately in order to know their risk. In the case of Pb, *H. scoparia, G. corneum* and *H. incurva* are the species that showed the highest results, with values of 5.7, 6.5 and 6.6 μg g^{-1} DW, respectively. These values could indicate a health hazard associated with the consumption of certain seaweed from the Basque Country, if compared to the hazard limits established by French regulations (5 μg g^{-1} DW). However, Rivas et al. (2022) showed that gentle depuration with tap water was effective in significantly reducing these elements. Furthermore, continuous monitoring heavy metals with high-risk potential in seaweed is deemed necessary to safeguard human health (Mendes et al. 2022).

In addition to the associated risks of metals, minerals have nutritional properties, such as iron (Fe), which is essential for almost all living organisms and plays a crucial role in a diverse range of metabolic processes (Abbaspour et al. 2014). Besides, Fe in seaweed is notably higher than in other vegetable sources (Norziah and Ching 2000) due to its metabolic system, which can directly absorb elements from the seawater. The report by Moreiras et al. (2013) identifies soybeans and lentils as legumes and vegetables with the highest iron content, providing 9.7 and 7.1 mg of Fe, respectively. These values are comparable to those found in the species studied (Table 3). In fact, concerning resource optimization, both *B. bifurcata* and *H. incurva* could serve as valuable new sources of Fe due to the high record they showed in the present study, posing seaweed could be considered a source of Fe and including it in our daily dietary food is feasible to increase Fe intake, as also García-Casal et al. (2007) described.

Furthermore, Mg is also an interesting mineral with beneficial nutritional properties, due to it serves as a cofactor in numerous enzymatic reactions and plays a crucial role in regulating biochemical pathways in the human body. However, although Mg is in high quantities in the studied species, especially for *U. rigida*, *H scoparia* and *C. acicularis*, the concentrations are significantly higher in many vegetables, whole grains, nuts and legumes.

In addition to Fe and Mg, other macroelements like Ca, K and Na, were present in high amounts for most of the studied species. K levels in the analyzed species (37,460.2 μg g^{-1} DW on average) were higher than those found in spinach (Rohani-Ghadikolaei et al. 2012). Besides, these high K concentrations are directly related to the ratio Na/K, which has nutritional significance, since a balanced Na/K ratio is crucial for hypertensive and K-deficient individuals. In the present study, the Na/K ratio ranges from 0.2 in *H. incurva* to 3.3 in *C. acicularis*, so including the studied species, which exhibit low Na/K ratios, in the diet could contribute to a balanced sodium–potassium ratio, offering an alternative to other foods with excessively high ratios(Kumar et al. 2011).

Protein composition

The results of this study are in good accordance with the literature, which suggests that proteins represent less than 15% of dry-weight in seaweed (D'Armas et al. 2019; Babich et al. 2022; Bass et al. 2023). Moreover, along with the present work, other studies (Fleurence 1999; Mateus et al. 2024), also indicate that Rhodophyta, compared to Chlorophyta and Phaeophyceae, is the richest in protein content, which can be attributed to the presence of PBP (Sanz et al. 2023), an exclusive pigment from red macroalgae, besides cryptomonads and cyanobacteria (Glazer 1994).

In this context, regarding resource optimization, while legumes, nuts, and animal protein origin generally exhibit the highest protein content, red seaweed could serve as an interesting alternative source. Specifically, as this study showed, *G. corneum, P. linearis,* and *H. incurva* contain protein levels of 13.45%, 16.27%, and 11.22%, respectively, which are comparable to the 12.7 g $(100\ g)^{-1}$ protein content of eggs or 11.7 g $(100\ g)^{-1}$ of oats (Ortega Anta et al. 2021). In this sense, incorporating seaweed proteins into our daily diet could offer a convenient option for easily obtaining these high-quality proteins (Cermeño et al. 2020). However, protein content varies considerably throughout the year depending on different factors such as the availability of nutrients (carbon, nitrogen and phosphorus), which bioavailability is regulated by environmental factors such as temperature, light, salinity, and water motion (Rawiwan et al. 2022). It is essential to identify which seasons or harvest places are most interesting for a given species and thus direct its use in the food industry (Samarathunga et al. 2022).

Ash content

As far as ash content is concerned, these results agree with the results obtained by other authors where ash content ranges from 19 to 56% (Sánchez-Machado et al. 2004; Neto et al. 2018). These amounts are generally much higher than terrestrial vegetables (Rupérez 2002; Sánchez-Machado et al. 2004). This high content could be explained by the accumulation of essential minerals, which can be due to the presence of anionic carboxyl, sulphate, and phosphate in their cell wall (Davis et al. 2003). The cell wall content

depends on the species and the month and harvesting areas (Samarasinghe et al. 2021; Premarathna et al. 2022), since they vary according on factors such as water composition and environmental conditions.

Conclusion

The macroalgal uses may be a promising alternative for many purposes, including, among others, pharmaceuticals, cosmetics, and food industries. However, variations in bioavailability and biochemical composition among species influence their suitability for different applications, highlighting the need for targeted research to optimize their potential for the different fields. In this study, none of the studied species produced adequate quantities of pigments to surpass other sources, except for phycobiliproteins, which could be extracted from *P. linearis* due to its good yield. Concerning microbiological analysis, washing the macroalgae (1 min under the tap water) and freezing (−20 ºC) affects microbial determinations, showing in both cases a generalized reduction (60–100%) of microorganisms. Hence, the use of both procedures is encouraged to reduce microbial levels, even when the limits allowed for human health are exceeded in fresh samples. In the analysis of minerals, Fe stands out for its high concentrations, surpassing typical high-Fe sources, making seaweed a potentially valuable source of human nutritional enrichment. Moreover, the concentration of both Hg and Cd metals does not exceed the recommended limits by European Regulations (European-Commission 19 March 2018; European-Comission 25 April 2023). However, the concentration of both As and Pb metals could show a potential risk for human use at higher consumption rates since the detected limits exceed the recommended limit by the French Food Safety Agency (CEVA 2020). Regarding proteins, the analysis highlighted the general elevated protein content, mainly in Rhodophyta, which emphasized the use of seaweed as a natural protein source for human consumption.

The assessment carried out herein provides new data on the main characteristics of common seaweed along the Basque Coast. However, it would be valuable to extend the analysis to seasonal and geographical contexts to explore additional potential variations. Furthermore, whereas some macroalgal species show great potential for some compounds, their large-scale extraction from the natural environment is neither sustainable nor viable. Therefore, it is also crucial to focus research efforts on developing methodologies for large-scale extraction and cultivation that ensure the continuous availability of algae biomass, focusing on the challenges of scaling up these processes, the target species and closing the gap between laboratory research and industrial implementation to achieve sustainable and commercially efficient production. In this context, considering the previous knowledge about their potential to be cultivated, this study identifies *P. linearis and C. acicularis* (Mouga et al. 2025), and the two studied Chlorophyta species as ideal candidates for cultivation in this area. Furthermore, in this sense, the use of seaweed as a source of bioactive and food/feed components is promising within the framework of the sustainable blue economy, not only due to the variety of key components that can be exploited, but also as a way to promote the CO_2 neutral processes in the industry.

Acknowledgements The authors express their gratitude to the NUMAPS (NUtrition MAnagement in Plant and Soil) group from the University of the Basque Country (UPV-EHU) for providing the use of some of their facilities for processing the samples.

Author contribution M.S.: Conception and design, Methodology, Analysis and interpretation of the data, Drafting and editing of the article. J.B. and A.A.: Methodology. N.E., E.Q., I.A., M.O., S.G-Z. and M.P.: Conception and design, and Methodology. S.S. and M.S.: Conception and design, Analysis and interpretation of the data, Project administration and funding acquisition and Supervision. All authors have critically reviewed the manuscript and have accepted responsibility for the entire content of this final manuscript.

Funding Open Access funding provided thanks to the CRUE-CSIC agreement with Springer Nature. This work was supported by the project 00002-INA2022-33 "SLOWALGA 3" funded by the University of the Basque Country (UPV/EHU), Spain, and the IT1471-22 and IT1743-22 projects consolidated research groups of the Basque Government.

Data availability The online version contains supplementary material available.

Declarations

Competing interest The authors declare no competing interests.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit http://creativecommons.org/licenses/by/4.0/.

References

Abbaspour N, Hurrell R, Kelishadi R (2014) Review on iron and its importance for human health. J Res Med Sci 19:164–174

Algaetraits (2024) AlgaeTraits: a trait database for (European) seaweeds. https://algaetraits.org; accessed 15 June 2024

Alisha, Dubey PR, Haider A (2019) Seaweed: Nutritional and health benefits. Pharm Innov 8:80–83
Andrade PB, Barbosa M, Matos RP, Lopes G, Vinholes J, Mouga T, Valentao P (2013) Valuable compounds in macroalgae extracts. Food Chem 138:1819–1828
Arriaga O, Wawrzynkowski P, Muguerza N, Díez I, Gorostiaga J, Quintano E, Becerro MA (2024) The thermal journey of macroalgae: Four decades of temperature-induced changes in the Southeastern Bay of Biscay. Mar Environ Res 195:106351
Atlas RM, Bartha R (1998) Microbial ecology: Fundamentals and applications. Benjamin-Cummings, NY, p 694
Babich O, Sukhikh S, Larina V, Kalashnikova O, Kashirskikh E, Prosekov A, Noskova S, Ivanova S, Fendri I, Smaoui S, Abdelkafi S, Michaud P, Dolganyuk V (2022) Algae: Study of edible and biologically active fractions, their properties and applications. Plants 11:780
Bartolomé L, Etxebarria N, Martínez-Arkarazo I, Raposo JC, Usobiaga A, Zuloaga O, Raingeard D, Cajaraville MP (2010) Distribution of organic microcontaminants, butyltins, and metals in mussels from the Estuary of Bilbao. Arch Environ Contam Toxicol 59:244–254
Bass AV, Smith KE, Smale DA (2023) Marine heatwaves and decreased light availability interact to erode the ecophysiological performance of habitat-forming kelp species. J Phycol 59:481–495
Beer S, Eshel A (1985) Determining phycoerythrin and phycocyanin concentrations in aqueous crude extracts of red algae. Mar Freshw Res 36:785–792
Borja A, Egaña J, Valencia V, Franco J (2000) 1947–1997, estudio y validación de una serie de datos diarios de temperatura del agua del mar en San Sebastián, procedente de su Aquarium. Ozeanografika 3:139–152
Borja A, Fontán A, Muxika I (2013) Interactions between climatic variables and human pressures upon a macroalgae population: Implications for management. Ocean Coast Man 76:85–95
Cermeño M, Kleekayai T, Amigo-Benavent M, Harnedy-Rothwell P, Fitzgerald RJ (2020) Current knowledge on the extraction, purification, identification, and validation of bioactive peptides from seaweed. J Electrophor 41:1694–1717
Ceva (2020) Edible seaweed and microalgae. Regulatory status in France and Europe. Centre d'Étude et de Valorisation des Algues France. CSHPF 01/1990
Chen K, Roca M (2018) In vitro bioavailability of chlorophyll pigments from edible seaweeds. J Funct Foods 41:25–33
Cherry P, O'hara C, Magee PJ, Mcsorley EM, Allsopp PJ (2019) Risks and benefits of consuming edible seaweeds. Nut Rev 77:307–329
Chust G, Borja A, Caballero A, Irigoien X, Sáenz J, Monjo R, Marcos M, Liria P, Hidalgo J, Valle M, Valencia V (2011) Climate change impacts on coastal and pelagic environments in the southeastern Bay of Biscay. Clim Res 48:307–332
Chust G, González M, Fontán A, Revilla M, Alvarez P, Santos M, Cotano U, Chifflet M, Borja A, Muxika I, Sagarminaga Y, Caballero A, De Santiago I, Epelde I, Liria P, Ibaibarriaga L, Garnier R, Franco J, Villarino E, Irigoien X, Fernandes-Salvador JA, Uriarte A, Esteban X, Orue-Echevarria D, Figueira T, Uriarte A (2022) Climate regime shifts and biodiversity redistribution in the Bay of Biscay. Sci Total Environ 803:149622
Cikos AM, Subaric D, Roje M, Babić J, Jerković I, Jokić S (2022) Recent advances on macroalgal pigments and their biological activities (2016–2021). Algal Res 65:102748
Cotas J, Leandro A, Pacheco D, Gonçalves AMM, Pereira L (2020) A comprehensive review of the nutraceutical and therapeutic applications of red seaweeds (Rhodophyta). Life Sci 10:19
Cruz G (2013) Boric acid in Kjeldahl analysis. J Chem Educ 90:1645–1648
Cunniff P (1995) Official Methods of Analysis of AOAC International. Association of Official Analytical Chemists, Washington DC, 1197 p
D'Armas H, Jaramillo C, D'Armas M, Echavarría A, Valverde P (2019) Proximate composition of several green, brown and red seaweeds from the coast of Ecuador. Rev Biol Trop 67:61–68
Davis TA, Volesky B, Mucci A (2003) A review of the biochemistry of heavy metal biosorption by brown algae. Water Res 37:4311–4330
Díez I, Muguerza N, Santolaria A, Ganzedo U, Gorostiaga JM (2012) Seaweed assemblage changes in the eastern Cantabrian Sea and their potential relationship to climate change. Estuar Coast Shelf Sci 99:108–120
Díez I, Santolaria A, Gorostiaga JM (2003) The relationship of environmental factors to the structure and distribution of subtidal seaweed vegetation of the western Basque coast (N Spain). Estuar Coast Shelf Sci 56:1041–1054
Echave J, Otero P, Garcia-Oliveira P, Munekata PES, Pateiro M, Lorenzo JM, Simal-Gandara J, Prieto MA (2022) Seaweed-derived proteins and peptides: promising marine bioactives. Antioxidants 11:176
Egan S, Harder T, Burke C, Steinberg P, Kjelleberg S, Thomas T (2013) The seaweed holobiont: understanding seaweed-bacteria interactions. Microb Rev 37:462–476
Esteban R, Martínez B, Fernández-Marín B, María Becerril J, García-Plazaola JI (2009) Carotenoid composition in Rhodophyta: insights into xanthophyll regulation in *Corallina elongata*. Eur J Phycol 44:221–230
European-Commission (19 March 2018) Recommendation (EU) 2018/464 on the monitoring of metals and iodine in seaweed, halophytes and products based on seaweed.
European-Comission (25 April 2023) Regulation (EU) No 2023/915 concerning maximum limits for certain contaminants in food. European Union**.** DOUE-L-2023–80614
Fleurence J (1999) Seaweed proteins: biochemical, nutritional aspects and potential uses. Trends Food Sci Technol 10:25–28
Freitas MV, Pacheco D, Cotas J, Mouga T, Afonso C, Pereira L (2022) Red seaweed pigments from a biotechnological perspective. Phycology 2:1–29
Galparsoro I, Borja Á, Legorburu I, Hernández C, Chust G, Liria P, Uriarte A (2010) Morphological characteristics of the Basque continental shelf (Bay of Biscay, northern Spain); their implications for integrated coastal zone management. Geomorphology 118:314–329
García-Casal MN, Pereira AC, Leets I, Ramírez J, Quiroga MF (2007) High iron content and bioavailability in humans from four species of marine algae. J Nutr 137:2691–2695
Glazer AN (1994) Phycobiliproteins - a family of valuable, widely used fluorophores. J Appl Phycol 6:105–112
Gorostiaga JM, Díez I, Casares C, Secilla A, De Castro A (2004) Check-list of the Basque coast benthic algae (North of Spain). Ann Jard Bot Madrid 61:155–180
Guiry MD, Guiry GM (2024) AlgaeBase. National University of Ireland, Galway. https://www.algaebase.org; accessed 15 June 2024
Hemlata FT (2009) Screening of cyanobacteria for phycobiliproteins and effect of different environmental stress on its yield. Bull Environ Contam Toxicol 83:509–515
Ioannou E, Roussis V (2009) Natural products from seaweeds. In: Osbourn A, Lanzotti V (eds) Plant Derived Natural Products. Springer, New York, pp 51–81
Jeffrey SW, Mantoura RFC, Bjornland T (1997) Data for the identification of 47 key phytoplankton pigments. In: Jeffrey SW, Mantoura RFC, Wright SW (eds) Phytoplankton pigments in oceanography: guidelines to modern methods. UNESCO, Paris, pp 149–559
Ji L, Qiu S, Wang Z, Zhao C, Tang B, Gao Z, Fan J (2023) Phycobiliproteins from algae: Current updates in sustainable production and applications in food and health. Food Res Int 167:112737
Kalvaitienė G, Vaičiūtė D, Bučas M, Gyraitė G, Kataržytė M (2023) Macrophytes and their wrack as a habitat for faecal indicator

bacteria and *Vibrio* in coastal marine environments. Mar Pollut Bull 194:115325
Kjeldahl J (1883) Neue methode zur bestimmung des stickstoffs in organischen Körpern. Z Anal Chem 22:366–382
Kumar M, Kumari P, Trivedi N, Shukla M, Gupta V, Reddy CRK, Jha B (2011) Minerals, PUFAs and antioxidant properties of some tropical seaweeds from Saurashtra coast of India. J Appl Phycol 23:797–810
Laurenns-Balparda Y, Seoane S (2024) Summer Ostreopsis blooms in San Sebastian (South-East Bay of Biscay): The importance of substrate features. Mar Pollut Bull 211:117484
Leong YK, Chen CY, Varjani S, Chang JS (2022) Producing fucoxanthin from algae - Recent advances in cultivation strategies and downstream processing. Bioresour Technol 344:126170
Løvdal T, Lunestad B, Myrmel M, Rosnes JT, Skipnes D (2021) Microbiological food safety of seaweeds. Foods 10:2719
MaM S, Hossain MS, Islam MM, Rahman M, Kibria G (2022) Heavy metals and metalloids in edible seaweeds of Saint Martin's Island, Bay of Bengal, and their potential health risks. Mar Pollut Bull 181:113866
Mateus M, Mão De Ferro R, Pinheiro HM, Machado R, Fonseca MMR, Sapatinha M, Pires C, Marmelo I, Marques A, Nunes ML, Motta C, Cesário MT (2024) Production and characterization of protein-rich extracts from the red macroalga *Gelidium corneum* and its industrial agar extraction residues. Algal Res 78:103420
Mendes M, Navalho S, Ferreira A, Paulino C, Figueiredo DR, Silva D, Gao F, Gama F, Bombo G, Jacinto R, Aveiro SS, Schulze PSC, Gonçalves AT, Pereira H, Gouveia L, Patarra RF, Abreu MH, Silva JT, Navalho J, Varela JCS, Speranza LG (2022) Algae as food in Europe: An overview of species diversity and their application. Foods 11:1871
Moreiras O, Carbajar A, Cabrera L, Cuadrado C (2013) Informe sobre legumbre, nutrición y salud. Tablas de composición de alimentos. Guía de prácticas, Fundación Española de la Nutrición, Madrid
Mouga T, Sousa A, Freitas MV, Afonso C (2025) Optimizing growth conditions and biochemical properties of *Chondracanthus acicularis* (Rhodophyta) in laboratory settings. Appl Sci 15:810
Muguerza N, Díez I, Quintano E, Bustamante M, Gorostiaga JM (2017) Structural impoverishment of the subtidal vegetation of southeastern Bay of Biscay from 1991 to 2013 in the context of climate change. J Sea Res 130:166–179
Munier M, Dumay J, Morançais M, Jaouen P, Fleurence J (2013) Variation in the biochemical composition of the edible seaweed *Grateloupia turuturu* Yamada harvested from two sampling sites on the Brittany Coast (France): The influence of storage method on the extraction of the seaweed pigment R-phycoerythrin. J Chem 2013:568548
Neto RT, Marçal C, Queirós AS, Abreu H, Silva AMS, Cardoso SM (2018) Screening of *Ulva rigida*, *Gracilaria* sp., *Fucus vesiculosus* and *Saccharina latissima* as functional ingredients. Int J Mol Sci 19:2987
Norziah MH, Ching CY (2000) Nutritional composition of edible seaweed *Gracilaria changgi*. Food Chem 68:69–76
Nowruzi B, Konur O, SaA A (2022) The stability of the phycobiliproteins in the adverse environmental conditions relevant to the food storage. Food Bioproc Technol 15:2646–2663
Nunes N, Valente S, Ferraz S, Barreto MC, Carvalho MaaPD (2020) Biochemical study of attached macroalgae from the Madeira Archipelago and beach-cast macroalgae from the Canary Islands: multivariate analysis to determine bioresource potential. Bot Mar 63:283–298
Ortega Anta R, López-Sobaler A, Andrés P, Aparicio A (2021) Composición nutricional de los alimentos. Nutrición y Ciencia de los alimentos, Universidad Complutense de Madrid, Herramienta para el diseño y valoración de alimentos y dietas
Osório C, Machado S, Peixoto J, Bessada S, Pimentel FB, Alves R, Oliveira MBPP (2020) Pigments content (Chlorophylls, fucoxanthin and phycobiliproteins) of different commercial dried algae. Separations 7:33
Pde (2024) Ministerio de transportes, movilidad y agenda urbana: Puertos del Estado. https://www.puertos.es/es-es/oceanografia/Paginas/portus.aspx; accessed 16 February 2024
Premarathna AD, Tuvikene R, Fernando PHP, Adhikari R, Perera MCN, Ranahewa TH, Howlader MM, Wangchuk P, Jayasooriya AP, Rajapakse RPVJ (2022) Comparative analysis of proximate compositions, mineral and functional chemical groups of 15 different seaweed species. Sci Rep 12:19610
Quintano E, Celis-Plá P, Martinez B, Díez I, Muguerza N, Lopez Figueroa F, Gorostiaga JM (2019) Ecophysiological responses of a threatened red alga to increased irradiance in an in situ transplant experiment. Mar Environ Res 144:166–177
Quintano E, Ganzedo U, Díez I, Figueroa F, Gorostiaga JM (2013) Solar radiation (PAR and UVA) and water temperature in relation to biochemical performance of *Gelidium corneum* (Gelidiales, Rhodophyta) in subtidal bottoms off the Basque coast. J Sea Res 83:106
Rawiwan P, Peng Y, Paramayuda I, Quek SY (2022) Red seaweed: A promising alternative protein source for global food sustainability. Trends Food Sci Technol 123:37–56
Rivas J, Piña F, Araya M, Latorre-Padilla N, Pinilla-Rojas B, Caroca S, Bronfman FC, Contreras-Porcia L (2022) Heavy metal depuration steps for *Gracilaria chilensis* in outdoor culture systems. Molecules 27:6832
Rohani-Ghadikolaei K, Abdulalian E, Ng W-K (2012) Evaluation of the proximate, fatty acid and mineral composition of representative green, brown and red seaweeds from the Persian Gulf of Iran as potential food and feed resources. J Food Sci Technol 49:774–780
Rupérez P (2002) Mineral content of edible marine seaweeds. Food Chem 79:23–26
Salido M, Soto M, Seoane S (2024) Seaweed: Nutritional and gastronomic perspective. A review. Algal Res 77:103357
Samarasinghe MB, Van Der Heide ME, Weisbjerg MR, Sehested J, Sloth JJ, Bruhn A, Vestergaard M, Nørgaard JV, Hernández-Castellano LE (2021) A descriptive chemical analysis of seaweeds, *Ulva* sp., *Saccharina latissima* and *Ascophyllum nodosum* harvested from Danish and Icelandic waters. Anim Feed Sci Technol 278:115005
Samarathunga J, Wijesekara I, Jayasinghe M (2022) Seaweed proteins as a novel protein alternative: Types, extractions, and functional food applications. Food Rev Int 39:4236–4261
Sánchez-Machado D, López-Cervantes J, Lopez-Hernandez J, Paseiro-Losada P (2004) Fatty acids, total lipid, protein and ash contents of processed edible seaweeds. Food Chem 85:439–444
Sanz V, Torres MD, Domínguez H, Pinto IS, Costa I, Guedes AC (2023) Seasonal and spatial compositional variation of the red algae *Mastocarpus stellatus* from the Northern coast of Portugal. J Appl Phycol 35:419–431
Schubert N, Garcia-Mendoza E, Pacheco-Ruiz I (2006) Carotenoid composition of marine red algae. Phycol J 42:1208–1216
Secilla A, Gorostiaga J, Díez I, De Castro A (1997) *Antithamnion amphigeneum* (Rhodophyta, Ceramiales) from the European Atlantic Waters. Bot Mar 40:329–332
Seoane S, Zapata M, Orive E (2009) Growth rates and pigment patterns of haptophytes isolated from estuarine waters. Sea Res 62:286–294
Shahnaz L, Shameel M (2009) Chemical composition and bioactivity of some benthic algae from Karachi Coast of Pakistan. Int J Algae 11:377–393
Spillias S, Cottrell RS, Kelly R, O'brien KR, Adams J, Bellgrove A, Kelly B, Kilpatrick C, Layton C, Macleod C, Roberts S, Stringer

D, Mcdonald-Madden E (2022) Expert perceptions of seaweed farming for sustainable development. J Clean Prod 368:133052

Thiviya P, Gamage A, Gama-Arachchige NS, Merah O, Madhujith T (2022) Seaweeds as a source of functional proteins. Phycology 2:216–243

Timoner I, Bosch-Collet J, Castell-Garralda V, Abuin S, Calderón J (2020) Estudio de la presencia de metales pesados y yodo en algas destinadas al consumo humano. Evaluación del riesgo asociado y su contribución a la dieta total. Departamento de Salud, Agencia Catalana de Seguridad Alimentaria

Van Den Hoek C, Breeman AM (1990) Seaweed biogeography of the North Atlantic: Where are we now? In: Garbary DJ, South GR (eds) Evolutionary Biogeography of the Marine Algae of the North Atlantic. Springer, Berlin, pp 55–86

Wells ML, Potin P, Craigie JS, Raven JA, Merchant SS, Helliwell KE, Smith AG, Camire ME, Brawley SH (2017) Algae as nutritional and functional food sources: Revisiting our understanding. J Appl Phycol 29:949–982

Zapata M, Rodríguez F, Garrido JL (2000) Separation of chlorophylls and carotenoids from marine phytoplankton: a new HPLC method using a reversed phase C8 column and pyridine-containing mobile phases. Mar Ecol Prog Ser 195:29–45

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Authors and Affiliations

Marina Salido[1,5] · Manu Soto[2,5] · Jone Bilbao[1,5] · Endika Quintano[1,5] · Néstor Etxebarria[3,5] · Inés Arana[4,5] · Maite Orruño[4,5] · Arkaitz Almaraz[4] · Saioa Gómez-Zorita[6,7,8] · María P. Portillo[6,7,8] · Sergio Seoane[1,5]

✉ Marina Salido
marina.salido@ehu.eus

Manu Soto
manu.soto@ehu.eus

Jone Bilbao
jone.bilbao@ehu.eus

Endika Quintano
endika.quintano@ehu.eus

Néstor Etxebarria
nestor.etxebarria@ehu.eus

Inés Arana
ines.arana@ehu.eus

Maite Orruño
maite.orruno@ehu.eus

Arkaitz Almaraz
arkaitz.almaraz@ehu.eus

Saioa Gómez-Zorita
saioa.gomez@ehu.eus

María P. Portillo
mariapuy.portillo@ehu.eus

Sergio Seoane
sergio.seoane@ehu.eus

[1] Plant Biology and Ecology Department, Faculty of Science and Technology, University of the Basque Country (UPV/EHU), Biscay, Spain

[2] Zoology and Animal Cell Biology Department, Faculty of Science and Technology, University of the Basque Country (UPV/EHU), Biscay, Spain

[3] Analytic Chemistry Department, Faculty of Science and Technology, University of the Basque Country (UPV/EHU), Biscay, Spain

[4] Immunology, Microbiology and Parasitology Department, Faculty of Science and Technology, University of the Basque Country (UPV/EHU), Biscay, Spain

[5] Research Centre for Experimental Marine Biology and Biotechnology (Plentzia Marine Station, PiE-UPV/EHU), Biscay, Spain

[6] Nutrition and Obesity Group, Department of Nutrition and Food Science, University of the Basque Country (UPV/EHU) and Lucio Lascaray Research Institute, Alava, Spain

[7] CIBEROBN Physiopathology of Obesity and Nutrition, Institute of Health Carlos III, Alava, Spain

[8] Bioaraba Health Research Institute, Alava, Spain