SEAWEEDS AS ALTERNATIVE PROTEIN SOURCES: DIGESTIBILITY AND

NANOSTRUCTURAL EVOLUTION UPON DIGESTION

MOTIVATION

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- ✓ Seaweeds are a potential source of alternative proteins, with interesting composition and many advantages over land-based biomass. However, they are still largely unexplored as food ingredients, mainly due to the lack of knowledge on their techno-functional and nutritional properties.
- ✓ The aim of this study was to investigate the potential of two different red seaweed species, typically used in the food industry for the extraction of agar, as dietary

protein sources. The impact of their distinct cell wall structure and different processing methods on the digestibility were also evaluated. **METHODOLOGY RESULTS FILMS SEAWEEDS EXTRUDATES** SEAWEEDS PROCESSING **RED SEAWEEDS** GC Gelidium corneum (GC_S) Gracilaropsis longissima (GL_S) Figure 2. AA profile of the native seaweeds and their processed products. The **protein content** was 15.5% for *G. corneum* and 15.6% for G. longissima. The proportion of essential AAs was 36% for G. corneum and 35% for G. longissima. The most abundant AAs in both seaweeds were Asp, Glu, Gly and Ala. Figure 1. Confocal microscopy images of the native seaweeds and their processed products. Proteins were stained with Processing affected the AA profile, with the Fast Green. proportion of more labile AAs, such as Lys, decreasing. G. corneum presented smaller and more compacted cell structures. Processing leads to cell wall distortion and outward migration of proteins. ■AVERAGE Extrudates • In vitro protein digestibility was low (<58%) for both seaweed (GC_E, GL_E) species, compared to other plant sources, such as pigeon peas **HOT PRESSING** (96%), black beans (82%) and peanuts (96%). 70 • In the case of *G. corneum*, the digestibility increased after each processing step, up to 62% for the films. 60 • In the case of G. longissima, the digestibility reached a maximum after the extrusion step (61%), being slightly reduced after further processing to obtain films (55%). The OPA method provided the largest errors, probably due to interference of coloured compounds from the seaweeds. 30 20 The highly resistant cell walls from G. corneum require Films (GC_F, GL_F) successive processing steps to be permeabilized and 10 promote the release of proteins. G. longissima cell walls are highly permeable, facilitating the release of proteins upon extrusion. Further IN VITRO PROTEIN DIGESTIBILITY GC_S GC_E GC_F GL_S GL_E GL_F processing leads to thermal degradation of some AAs, Figure 3. Protein digestibility obtained from TN, TAA and OPA and the average values from the three having a negative impact on the digestibility. methods. In vitro Lysine is highly susceptible to thermal processing, Total digestibility ntestinal endpoint DIAAS becoming the limiting AA in the processed materials. Food GL_F GC_S GC_E GC_F GL_S GL E TN R-NH₂ TAA **Lowest DIAAS** 25.03 68.67 25.49 23.94 20.12 20.87 Supernatant **Limiting AA** LYS LYS SAA LYS LYS HIS Sample before digestion Hydrolysis 6 N HCI, 110°C, 15 h Pellets → non-digested Precipitation Digest Pellet Kjeldahl HPLC OPA material Nanostructure of the digestion products Digest Supernatant Pellet Nanostructural changes Control 2 Hydrolysis 6 N HCI, 110°C, 15 h occur only after **HPLC** Kjeldahl extrusion+hot pressing GELIDIUM C. GELIDIUM C. GELIDIUM C. $\frac{1}{(Fs-Cs)+\max(0;Fp-Cp)}*100$ Digestibility[%] = cookie **SEAWEED EXTRUDATE FILM** Sousa, R. et al. Food Chemistry, 404, 134720 (2023) Two types of structures В detected in supernatants: NANOSTRUCTURAL CHARACTERISATION OF THE **DIGESTION PRODUCTS** BS mixed lamellae A В **DIGESTS** supernatant ---- Indo film ----- Indo film pellet ----- Indo film superna Indo extr Indo extr pellet Indo extr supernat GRACILARIA L. GRACILARIA L. GRACILARIA L. **SEAWEED EXTRUDATE** FILM Vesicles pellet В **SMALL ANGLE X-RAY** A SCATTERING (SAXS)

CONCLUSIONS

Figure 4. SAXS patterns of the digests from the seaweeds and their processed products.

- The two studied red seaweed species contain relatively high protein contents and high proportion of EAAs. However, their tough cell walls limit protein diffusion. This, together with the presence of gelling polysaccharides such as agar, hinder protein digestibility. Thus, native seaweeds present lower protein digestibility than other plant sources.
- Processing through conventional methods such as extrusion and hot pressing induces cell wall distortion and permeabilization, hence increasing protein digestibility.
- Cell wall structure and composition are key to determine protein digestibility of native seaweeds and the effect of processing.
- Protein digestion products can interact with components from the physiological medium to form different types of nanostructures. This is expected to have a strong impact on intestinal transport.



