



Are Algal Proteins a Viable Alternative to Animal Protein? - A Review

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Abstract

Protein is a macronutrient extremely important for human health. This macronutrient presents a wide variety of biological functions - structural, catalytic, storage, signaling, protective - and, therefore, it is essential that individuals include it properly in their dietary patterns. Nonetheless, the extreme dependency on livestock to fulfill human requirements has a strong negative impact on the environment. Future predictions point to an increase of the global population from 7.7 billion to 11.2 billion by the end of the century and not enough alternative and efficient solutions to overcome this problem.

Algae represent a promising source since, when compared to vegetal sources, they have higher growth and photosynthesis rates. Besides, their potential to produce bioactive compounds that can be included in co-production with protein and the dispensability of arable land for their cultivation make them excellent candidates to support this demand.

This article aims to present a review of the scientific literature on the protein algal properties and their ability to replace animal protein.

Keywords: Protein; Algae; Food Sustainability; PDCAAS

Abbreviations

DIAAS: Digestible Indispensable Amino Acid Score; EAA: Essential Amino Acids; FAO: Food and Agriculture Organization of the United Nations; GHG: Greenhouse gases; IOM: Institute of Medicine; PDCAAS: Protein Digestibility-Corrected Amino Acid Score; PEF: Pulsed Electric Field; PUFA: Polyunsaturated Fatty Acids; RDA: Recommended Dietary Allowances; USDA: United States Department of Agriculture; WHO: World Health Organization

Introduction

Protein is a macronutrient that has a very important physiological role [1]. Whole-body protein balance has a strong

impact on muscle mass, function, bone and calcium homeostasis, immune system response, fluid and electrolyte balance and in enzymatic and hormonal systems [2]. In addition to this, high-protein diets may reduce overweight and obesity, one of the biggest diseases of this century [3]. In fact, following higher-protein diets in contrast to lower protein diets result in greater weight loss, waist circumference, fat mass loss, better preservation of lean body mass, reduction of blood pressure and serum triglycerides [4] and a lower developing cardiometabolic disease [5]. Besides, protein is known as the most satiating macronutrient [6].

The Institute of Medicine (IOM) and World Health Organization (WHO) defined that Recommended Dietary Allowance (RDA) for both adult men and adult women is 0,8 g/kg/d although higher

intakes may be required when adults are purposefully stressed by the catabolic stimulus of energy restriction or the anabolic effects of resistance training [7]. In order to meet these requirements and attending the global population growth projection to 9.5 billion by 2050, it is crucial to find solutions, concerning world sustainability and food security, since it is generally accepted animal-based foods produce higher levels of greenhouse gases (GHG) [8].

Impact of animal-based foods in environment

According to the Food and Agriculture Organization of the United Nations (FAO), 56 million land animals are raised and killed every year for human consumption; this sector represents 18% of GHG emissions, more than all transport worldwide [9]. Most emissions related to this industry are in the form of carbon dioxide, a significant contributor to global warming and climate change, nitrous oxide, ammonia and methane [10]. The last one has a 23-fold greater potential for global warming than carbon dioxide and, globally, about 80 million tons of methane is produced annually from enteric fermentation from livestock [11]. Besides, livestock is responsible for almost 64% of total ammonia emissions, contributing to acidification of ecosystems.

Furthermore, 30% of the Earth's land is involved in livestock production, contributing to very important environmental issues, such as land degradation, water pollution, overgrazing and desertification. According to the United States Department of Agriculture (USDA) animal parts and poultry represent major sources of water pollution [9], due to animal excreta, antibiotics, hormones fertilizers and pesticides used in forage production and rainfall runoff from grazing [10].

It is crucial to minimize the adverse environmental issues caused by animal-based foods and the incorporation of healthy and sustainable sourced plant proteins might be the solution [6]. Indeed, according to Ruini, *et al.* in one day, a vegan person saves 4164 L of water, 20 Kg crops, 2.8 m² of forested land, 10 Kg carbon dioxide and the life of one animal [12].

Characteristics of macro- and microalgae

Marine plants, such as macro- and microalgae, represent a promising protein source. The term *algae* embrace a group of mostly photosynthetic organisms with simple reproductive structures [13]. *Algae* are, generally, defined as eukaryotic organisms and contain valuable compounds with variable

concentrations according to their species and growth conditions [14]. They contain about 60% dry weight as protein. Although both are termed *algae*, there are differences between macro- and microalgae [8]. Macroalgae, often fast-growing and reaching sizes up to 60 m in length, are multicellular organisms and microalgae are unicellular, measuring 1 µm to several cm [13]. A standard classification has been established dividing macroalgae based on their pigmentation - brown seaweed (*Phaeophyceae*), red seaweed (*Rhodophyceae*) and green seaweed (*Chlorophyceae*). Microalgae include diatoms (*Bacillariophyceae*), green algae (*Chlorophyceae*), golden algae (*Chrysophyceae*) and blue-green algae (*Cyanophyceae*) and they, normally, grow in seawater in the photic zone, up to a depth of 200 m.

Macroalgae, or seaweed, can be cultivated in seawater and, generally, does not compete for arable land or freshwater [15], which makes it a potential candidate to a viable alternative to traditional agriculture and animal protein, contributing to a reduced carbon footprint and mitigating climate change. Furthermore, seaweed farming does not need pest- insect- or fungicides and provides habitats for fish and crayfish species and sequestering nutrients [16]. Recent findings also suggest phosphorus and carbon uptake from seaweed cultivation can contribute to mitigating coastal eutrophication and ocean acidification, respectively. Although its under-exploitation in food terms in the occidental world, most of the seaweeds are edible [15]. Their high content in dietary fiber, mineral, vitamin and phytochemical content, low energy levels and high concentrations of certain polyunsaturated fatty acids (PUFA) turns them in a very interesting food group [17].

Microalgae are microscopic bodies that grow in suspension in seawater, in the region where the sun penetrates, or in bioreactors, and its metabolites have been studied these past years. Several bioactive compounds have been found there, such as carotenoids, omega-3 fatty acids and polyphenols [18]. Microalgae, as macroalgae, has enormous potential as a part of the human diet, with their antioxidant, anti-inflammatory, anticancer and antiviral properties.

Nonetheless, the widespread use of seaweed and microalgae is limited by many factors, such as harvesting licenses, seasonality, geography and scalable production methods [19]. Indeed, there are emergent challenges associated with *algae*, including the warranty of the production sustainability.

Quality of vegetal protein: the case of algae

Dietary protein quality can be defined as several things, such as protein to support optimal growth, amino acid balance, indispensable amino acid related to essential amino acids (EAA) requirements and extent of digestion and absorption by the human body. According to Berrazaga, *et al.* protein quality depends on the EAA composition and the ability of the protein to be digested, absorbed and retained for the body [20].

There are nine EAA - histidine (His), leucine (Leu), isoleucine (Ile), lysine (Lys), valine (Val), methionine (Met), phenylalanine (Phe), tryptophane (Trp) and threonine (Thr) [21]. The nutritional value of dietary protein is directly related to these amino acids' bioavailability; the higher the efficiency of their metabolic utilization to meet amino acid human requirements, the better the quality.

Several methods can be used to evaluate the quality of a certain protein source, although the most accepted are the Protein Digestibility-Corrected Amino Acid Score (PDCAAS) and Digestible Indispensable Amino Acid Score (DIAAS) [22]. These can relate EEA content of an individual foodstuff to a reference EEA profile calculated to meet EEA requirements (Table 1) for each populational group and corrected for protein or EEA digestibility.

	EAA Requirement for adults								
	Lys	Leu	Ile	Val	Thr	Trp	Met	Phe + Tyr	His
mg/kg per day	30	39	20	26	15	4	10	25	10

Table 1: Adult essential amino acid requirements. Adapted from FAO, 2007.

PDCAAS can be calculated through the product of the Amino Acid Score (AAS) and the percent true fecal protein digestibility of the foodstuff which is being analyzed [23]. The AAS reflects the most limiting amino acid supplied in the foodstuff in comparison with a reference requirement pattern (Table 2).

The lowest calculated AA ratio is considered the limiting amino acid and used in the formula to estimate PDCAAS, as shown below. True protein digestibility is determined through the amount of fecal nitrogen excreted per unit of dietary nitrogen consumed.

$$PDCAAS (\%) = \frac{\text{mg of limiting amino acid in 1 g of test protein}}{\text{mg of same amino acid in 1 g of reference protein} \times \text{fecal true digestibility} (\%) \times 100}$$

Reference Pattern (mg/g of protein)								
Lys	Leu	Ile	Val	Thr	Trp	Met+Cys	Phe+Tyr	His
45	59	30	39	23	6	22	38	15

Table 2: Recommended FAO/WHO reference pattern. Adapted from Dupont, *et al.* 2020.

Usually, for the different plant sources, their total EEA content is lower than in animal food-based protein, such as meat, eggs and milk [24].

Also, the digestibility is lower for protein plant sources which might be explained by their different structures. Indeed, the secondary structure of plants is characterized by having a high content of β-sheet conformation and low content in α-helix conformation, unlike animal protein [20], which is related to resistance to proteolysis in the gastrointestinal tract. Aside from this, antinutritional compounds, non-starch polysaccharides and fibers typically present in plants can avoid the access of enzymes to proteins or interfere in its activity, decreasing its digestibility.

Indeed, all these facts contribute to lower PDCAAS in plant-based protein sources, as shown in table 3.

Source	PDCAAS
Milk	1
Whey	1
Egg	1
Soy protein isolate	1
Casein	1
Beef	0.92
Soy	0.91
Black bean	0.75
Pea	0.67
Oat	0.57
White rice	0.56
Peanuts	0.52
Whole wheat	0.45

Table 3: PDCAAS of some protein sources. Adapted from Van Vliet, *et al.* 2015 and Berrazaga, *et al.* 2019.

A dietary protein cannot fulfill human requirements if its PDCAAS is lower than 100 [20]. The plant-based protein sources have been shown to have a PDCAAS lower than 100% and, therefore, lower than animal-based protein sources, compromising human protein synthesis.

As well, plant protein has a lower content of leucine, the most potent amino acid responsible for muscle protein synthesis. Hence, anabolic properties from this type of dietary protein are limited and can only be overcome with greater amounts of plant-based proteins (< 30g/meal) or fortification with the limiting amino acid [24].

Consequently, plant proteins are, generally, considered incomplete as they lack one or more EEA and for being less digestible rather than animal protein sources [19].

Nonetheless, algae can be considered a viable protein source, meeting EEA requirements [18]. Indeed, microalgae amino acid profiles are well-balanced and similar to high-quality sources, such as lactoglobulin and egg albumin. Tryptophane and lysine are the most common limiting amino acids in algae, although different species of algae can differ in the limiting amino acids. For instance, in brown algae species, the most usual limiting amino acids are methionine, cysteine and lysine and in red species algae, leucine and isoleucine are also found in low concentrations. Among marine macroalgae, protein content and amino acid balance can have seasonal changes so the harvesting should occur when the protein content is favorable [25].

In what bioavailability is concerned, it is important to analyze both concepts which are englobed by bioavailability [25]. The first one is bioaccessibility and it refers to the release from the food matrix, transformations through digestive processes and transport along the digestive system and the second one is bioactivity which refers to the uptake of the nutrients into tissues, the metabolism and physiological effects. Due to its ethical and practical issues, studies on algae' bioactivity are based on short-term *in vitro* tests which can lead to a lack of information on the behavior of algae in the human body. Analytical methods, such as, simulated gastrointestinal digestion and genetic techniques could help to assess algae' bioavailability; nonetheless, its use is very limited in this food group. Consequently, current knowledge on functional

and nutritional algal value is limited. It is extremely important to develop research on micro- and macroalgae protein bioavailability, incorporating PDCAAS estimations.

Nonetheless, algae proteins seem to be digested less completely than casein *in vitro* model systems due to inhibitory soluble fibers, although also here the research is short. Pre-analytical steps such as freezing, milling, digesting the sample with enzymes, osmotic rupture and cooking should be a focus of research to improve algae' digestibility.

Protein extraction methods

Macro- and microalgae have poor protein digestibility in their unprocessed form and current literature suggests that the protein extraction method has a strong impact on its digestibility [19]. Thus, it is necessary to develop protein extraction methods that might improve protein bioavailability.

To obtain a functional protein extract, in dried or concentrated form, a first cell disruption is required, since the most of protein content is inside the cell [26]. Then, the process is followed by the protein solubilization/fractionation, purification and concentration. If the protein extraction process is well designed, pigments contents can be reduced and taste improved, solving two of the major problems of algae usage in the food industry.

For cell disruption, it can be used mechanical shear (high pressure homogenization, bead milling and ultrasonication), chemical/enzymatic hydrolysis and external fields, such as pulsed electric field (PEF). Although high-pressure homogenization has been suggested as the most efficient process in terms of protein release, other methods have brought the researcher's attention. Ultrasonication and chemical lysis have only been shown effective at very high energy inputs, leading to the degradation of the protein. The same happens with enzymatic treatments - since microalgae and cyanobacteria have very complex cell walls, the amount and mixture of enzymes could affect the functional properties of proteins.

In what PEF is concerned, it does not lead to a higher protein release due to microalgae rigid cell wall structures; nonetheless, pulsed electric fields do not completely disintegrate the cell wall, enabling the diffusion of macromolecules outside the cell

and, therefore, there is a lower amount of pigments that are co-extracted. This is a huge vantage for PEF usage, although its lower and slower protein extraction. Furthermore, there is evidence that in some PEF conditions, it is possible an efficient protein extraction along with the microalgae culture survival [27], and, therefore, it is possible to establish a protocol for continuous protein extraction during microalgae cultivation [28].

Conclusion

Although algae might be a promising resource to meet human demand, current cultivation systems are not yet competitive in comparison with other resources. Growth stimulation and selective microbial inactivation through nanosecond pulsed electric field (nPES) technology could be one way to increase cultivation performance; however, applying PEF on a large-scale is not yet viable and requires further investigation. Also, more research is required in what PEF variables are ideal to protein extraction.

Also, the application of microalgae proteins has been limited in foods mainly due to the presence of non-protein components that might affect the color and the taste of the final product. Besides, the rigid cell wall of some strains can lower digestibility.

Another issue is linked to safety hazards, which should be a topic of concern. It exists potential accumulation of heavy metals, high levels of iodine and contaminants that can compromise human's health. Indeed, there are very tight regulatory restrictions on novel foods, food safety and health claims. All these restraints might delay the commercialization of algae products.

Nutritionally, algae protein can be an effective alternative to animal protein. Nonetheless, it is mandatory to further research on algae bioavailability and interactions inside the human body.

Conflict of Interest

The author has no conflicts of interest to declare.

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