



Research Paper

## Green seaweed, *Ulva* sp. a potential source of ingredients for feed formulation of Ornamental fishes

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**ABSTRACT:** Seaweed is an admirable source of protein, dietary fiber, agar, alginate, carrageenan and , vitamin. *Ulva lactuca* is a rich source of protein (15-20%) and Carbohydrate (42–46%); it plays a vital role in increasing the growth of fish due to its rich nutrients. Cost effective resources such as seaweed could be suitably used as feed ingredients to culture freshwater ornamental fish. It also contains total carotenoids (0.5 to 1.5% in dry weight) which could increase pigmentation in the skin of ornamental fishes. Carotenoids like antioxidants help prevent the free radicals' damage associated with the aging process itself. This seaweed with rich bioactive compounds is also utilized in Medical field in the formulation of drugs for humans. Similarly, it could be used as a drug against fish disease. This seaweed is also used as pellet binder in fish diets and as partial substitution of rich protein sources in animal feeds. Fish feed cost is one of the major input tasks in the fast growing field of ornamental fish culture and for the growth of the industry. Hence, this review will provide a new dimension output and support using *Ulva lactuca* as a partial ingredient of feed.

**KEY WORDS:** *Ulva lactuca*, Carbohydrates, Carotenoids.

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### I. INTRODUCTION

There is sustained interest in exploring the aqua ingredients and fish feeds but much remains unknown about the usage of aquatic macroalgae (seaweeds) in nutrition of aquatic organisms. Cost effective and naturally colourful feeds using the available seaweeds can be prepared in our country. Cyanobacteria (blue green algae such as *Spirulina*), dried shrimp meal, shrimp and palm oils, extracts from marigold, red peppers and *Phaffia* yeast are excellent natural sources of pigments. Antioxidants, in particular carotenoids, help prevent the free radicals' damage associated with the aging process itself. Epidemiological evidence strongly suggests that food rich in carotenes or vitamin A are associated with a reduced health risk which happened when seaweed was used as an ingredient for ornamental fish. Most fish have powerful sensory characters. If the flavour and odour of food are not good they reject the feed. The characteristic of food is very important especially for bottom feeders. Flavour and taste, sound, smell, colour and buoyancy of food are the main factors that influence the feeding of ornamental fishes. The aim of this review paper is to evaluate the nutritional value and the use of seaweeds as pellet binder in ornamental fish diet.

#### Seaweeds and their nutritional importance

The genus *Ulva* (Phylum: Chlorophyta, Class: Ulvophyceae, Order: Ulvales, Family: Ulvaceae) was first identified by [1] and [2]. About 20,000 species found distributed globally, cosmopolitan and important wealth for their food, pharmaceutical and nutritional values. It also could be used as a nonconventional source for animal feeding which may contribute to solving environmental problems [3]. It is a commercially important marine living and renewable resource in India. The seaweed culture is a growing worldwide industry; produced 14.5 million tones (wet weight) worth US\$7.54 billion in 2007 [4]. Seaweeds have been used as animal feed since the first century BC by the Greek's. Seaweeds have also been used for many years in human nutrition;

largely as a vitamin additive and feeding stuff for ruminants [5] and [6]. This explains why today in 21st century seaweeds are considered as the food supplement as source for proteins, lipids, polysaccharides, mineral, vitamins and enzyme [7]. They contain more than 60 trace elements, minerals, protein, iodine, bromine, vitamins and several bioactive substances of economic value [8]. The best known components of the seaweed-derived industry are: phycocolloids, the gelling, thickening, emulsifying, binding, stabilizing, clarifying and protecting agents known as carrageenans, alginates and agars [9].

Seaweed is an excellent source of protein, dietary fiber, alginate (thickener in drinks, ice cream) agar, carrageenan (thickener and clarifier), vitamins (especially A, B1, C, and E) iron and iodine. *Ulva lactuca* contains natural carotenoids, so it prevents the addition of artificial colouring agents. Carotenoids are an important group of natural pigments with specific applications as colorants, feed supplements. Despite the availability of a variety of natural and synthetic carotenoids, only a few have been exploited commercially, including  $\beta$ -carotene, lycopene, astaxanthin, canthaxanthin, lutein and capxanthin [10]. Antioxidants in particular carotenoids, help prevent the free radicals' damage associated with the aging process itself. The review by the National research council (NRC, 1982) concluded "the epidemiological evidence sufficient to suggest that food rich in carotenes or vitamin A are associated with a reduced risk of cancer". Thus foods rich in carotenoids, in this case  $\beta$ -carotene, may not only be able to prevent but also reverse cancers. A lot of studies demonstrated the antioxidant properties of algal carotenoids and the role they play in preventing many diseases linked to oxidative stress [11], especially trace elements and several other bioactive substances.

[12] reported that the crude protein content of *Ulva sp.* ranged between 10 and 26% of dry Weight. Only few studies have been undertaken on the quality of seaweed proteins because the extraction of protein from seaweed is difficult due to presence of phenolic compounds and large amounts of polyanionic cell wall mucilages [13] and [14].

In ornamental fish trade, freshwater ornamental fish contribute 85 per cent of the total and the rest are from marine species. Though Asian countries contribute 90 per cent of the ornamental fish trade around the globe, India's share to the global trade is less than 0.01 per cent, till recently. But the ornamental fish export in India is growing at the rate of 10 percent per annum during the last few years. At the same time, the domestic ornamental fish market in the country is also growing at the rate of 20 per cent per year. India is very rich in terms of ornamental fish genetic resources with more than 200 varieties of indigenous ornamental fish available in the North Eastern States and Western Ghats of the country [15]. At present, ornamental fish farming is the fastest growing aquaculture sector in the world with a global trade of US \$ 9.0 billion per annum and a growth rate of over 8 per cent. Although ornamental fish farming has already been recognized as the significant contributor to the Indian aquaculture industry, the main problem in the growth of this industry is the non-availability of quality feed at an affordable price.

Many of the ornamental fish growers until now grow the ornamental fish using the diets that are meant for other food fish species. Moreover, the ornamental fish industry in India is fully dependent on the imported feeds, which are not only costly but also not sure about their nutrient quality. [16] reported that the crude protein content of *Ulva sp.* ranged between 10 and 26% of dry weight [17]. This protein is considered high quality protein, since the green algae, *Ulva lactuca* contains all the essential amino acids (EAA) and accounted for 42.1-48.4% of the total amino acids content except tryptophan [18]. This could be used as a feed supplement [19]. About 100g wet weight of this seaweed contains: total fiber: 3.8, soluble fiber: 2.1, insoluble fiber: 1.7, Carbohydrates: 4.1, calcium: 325.0, potassium: 245.0, Magnesium: 465.0, Sodium : 340.0, Copper: 0.3, Iron: 15.3, Iodine: 1.6 and Zinc: 0.9. Thus, cost effective nutrients are present in different seaweeds.

[20] stated that 100g dry matter of *Ulva sp.* contains vitamins A, B1, B2, B3, B12, folic acid and C at the rate of 960 IU, 0.06, 0.03, 8.0, 6.3, 11.8 mg and 10.0 mg, respectively. It is worth mentioning green algae as an important source of Polyunsaturated Fatty Acids (PUFAs). Due to nutritional values and beneficial effects of PUFAs, many researchers have investigated the suitability of using macroalgae as novel dietary sources of PUFAs. [21] have detected higher percentages (16%) of  $\alpha$ -Linolenic Acid (ALA) in *Ulva sp.*, collected at the Algarve coast, Portugal.

Seaweeds act as diets to realize better growth and dietary performance of ornamental fish, for which the determination of nutrient requirements is the basic prerequisite. The nutritional value of such supplements is generally evaluated in terms of growth and survival, with little attention paid to other physiological merits. Use of dietary algae as feed supplements may be expected to improve growth and digestive efficiency of feed [22]. The addition of small amounts of algae to the fish diet can produce considerable improvement in physiological condition, fish vitality, disease resistance, desired body composition and carcass quality [23].

Feed is the major input to ornamental fish culture because of the increased habit of keeping aquariums. Currently, the feed cost is increased due to demand; to meet this demand the cost effective quality feed is essential to produce high health and profitable fishes. Over feeding and leaching feed ingredients also lead to water pollution and health risk. Essential or indispensable amino acids (EAAs) cannot be synthesized by fish

and often remain adequate, but are needed for growth and tissue development [24] and [25]. Hence, to improve the fish health and water quality, cost effective quality protein source is essential. It is important to mention also that ulvan, a major Sulfated Polysaccharide (SP) found in the cell wall of green algae, composed mainly of rhamnose, glucuronic acid, iduronic acid, xylose, glucose, sulphate with small amounts of iduronic acid and traces of galactose and represents 8-29% of the algae dry weight [26] and [27]. The presence of the sulfate groups and the unusual chemical composition and structure of *Ulva sp.* render it different biological properties [28], [29] and [30].

### Characteristics of Seaweed ingredients Carbohydrate

Carbohydrate serves as the least expensive source of dietary energy and helps in improving the pelleting quality of practical fish diets [53]. Feed cost per unit of fish produced can be minimized by optimal use of low-cost energy carriers such as carbohydrate-rich ingredients, ensuring that the use of costly protein is kept as low as possible [54]. Replacing dietary protein by carbohydrate or lipid energy may result in a higher production per unit spent on costly protein sources such as fishmeal, and the effluent nitrogen can be reduced per unit of fish produced [31]. Carbohydrates also served as precursors for various metabolic intermediates necessary for growth. *Ulva reticulata* which contains 55.77 (g/100g sample dry basis). We used these green seaweeds as fish ingredients to give the proper growth of fish. The CHO are involved in the secretion and activity of insulin and glucagon and in less proportion in the growth hormones [32].

### Protein and Amino acids

[33] indicated that although the Green seaweed (*Ulva lactuca*) proteins contained all essential amino acids, the levels of which were comparable to those of the FAO/WHO requirement. For *Ulva reticulata*, the protein was three times higher than that contained in the same genus *Ulva lactuca* but slightly lower than that of *Porphyra sp.* [34]. However, it should be noted that the protein content of seaweeds varied not only between species but also between seasons [35]. However, [36] reported that the crude protein content of *Ulva sp.* ranged between 10 and 26% of dry weight which imply the potential for animal nutrition (e.g. as functional food and fish feed). This protein is considered high quality protein, since the green algae (e.g. *Ulva lactuca*) contain all the essential amino acids (EAA) [37]. *Ulva sp.* which have the important Essential or indispensable amino acids (EAAs) cannot be synthesized by fish and often remain inadequate but are needed for growth and tissue development [38]. However, [39] showed that green algae (*Ulva lactuca*) contain all the essential amino acids except tryptophan. *U. lactuca* contained 12% amino acids in which essential amino acids constituted 5%. The ratio of essential amino acids to total amino acids was 0.45 and that of nine contents was higher than those in all other seaweeds. *Ulva reticulata* proteins were of high quality because the essential amino acids represented almost 40% of total amino acids and the essential amino acids profile were closed to those of egg and soya protein [40], except for relative lack of data on tryptophan, methionine and cysteine.

### Lipids and fatty acids:

As the total lipids content of seaweeds was quite low so they were not a conventional source of energy. However, most of them were reported to be rich in polyunsaturated fatty acid regarding their fatty acid composition [41]. Variations in fatty acid contents are due to both environment and genetic differences mentioned above Sanchez-[42]. Fatty acid in *Ulva reticulata* is C16:0 (palmitic acid), which accounted for 41.53% for *Ulva reticulata* [55], [56] and [57]. However, they also contained the essential fatty acids of C18:2(w-6) (linoleic acid), C18:3(w-3)(alpha-linolenic acid), C20:5(w-3) (the eicosanoid precursors), C20:4 (w-6) (arachidonic acid) and C20:5 (w-3) (eicosapentaenoic acid) in rather small amounts. Moreover, PUFAs can be considered also as having an importance. [43] has reported that the Hexadeca-4, 7, 10, 13-Tetraenoic Acid (HDTA), Octadeca-6,9,12,15-Tetraenoic Acid (ODTA), and  $\alpha$ -Linolenic Acid, isolated from the methanol extract of *U. fasciata* by bioassay-guided fractionation [58] and [59].

**Table:1 Biological activities of *Ulva sp.***

Species	Extracts or Metabolites	Biological activities	References
<i>U. compressa</i>	Methanol	Antioxidant	Paiva LS,2012
<i>U. conglobata</i>	Sulfated polysaccharides	Anticoagulant	Mao W, Zang X, Li Y, Zhang H(2006)
<i>U. lactuca</i>	Dichloromethane/methanol	Antimicrobial	El-Baky HHA, El-Baz FK, El-Baroty GS (2009)
	Ethanol	Antimicrobial, antiparasitic	Kim KM, Hwang IK, Boo JK (2011), Lalvani A, et al. (2010)
	Dichloromethane/methanol	Antioxidant	El-Baky HHA, El-Baz FK, El-Baroty GS (2009)

	Methanol	Antitumor, antimicrobial, antiviral, and immune stimulating	Lee S-H, et al. (2004)
	Sulfated polysaccharide	Inhibits Japanese Encephalitis Virus (JEV)	Chiu YH, Chan YL, Li TL, Wu CJ (2012)
	3-O-β-D-glucopyranosylstigmasta-5,25-diene (6)	Anti-inflammatory	Awad NE (2000), Costa SS, et al. (2010)
	Methanol	Anti-inflammatory	Margret RJ, Kumaresan S, Ravikumar S (2009)
<i>U. fasciata</i>	Dichloromethane/methanol	Antiviral	Costa SS, et al. (2010)
	N-palmitoyl-2-amino-1,3,4,5-tetrahydroxyoctadecane (8)	Antiviral	Gamal AAE (2010)
	Dichloromethane/methanol	Antibacterial	Selvin J, Lipton AP (2004)
<i>U. fasciata Delile</i>	Ethanol	Anti proliferative	Chung HS, Kim H, et al. (2013)
	Labda-14-ene-3α,8α-diol(13) and Labda-14-ene-8α-hydroxy-3-one(15)	Antimicrobial	Chakraborty K, Lipton AP, Paulraj R, Vijayan KK (2010)
	Methanol	Antioxidant	Chakraborty K, Paulraj R (2010)
<i>U. rigida</i>	Diethyl ether and methanol	Antimicrobial	Tüney I, Çadirci BH, Unal D, Sukatar A (2006)
	Dichloromethane and ethanol	Cytotoxicity	Cunningham G, et al. (1999)
	Ethanol Antimutagenic,	antioxidant	Bagher N, et al. (2012); Silva <i>et al.</i> , 2013.

**Citation:** Silva *et al.*, 2013.

### Vitamins

Vitamins are necessary in the diet for normal fish growth and health. These are organic substances that act as catalysts for many of the biochemical reactions within the fish. They often are not synthesized by fish, and must be supplied in the diet [43] and [60]. Vitamins play a part in processes which are related to stress responses and the immune system, as they act as cofactors of enzymes, as antioxidants and as structural components of phospholipids [44]. Some important function of vitamin is listed as Thiamine is essential for a good appetite, normal digestion, growth, and fertility [61], Riboflavin function as enzymes of tissue respiration, Pyridoxine(B6) plays a vital role in enzyme systems and protein metabolism, panthotenic acid(B5) is essential for the development of the central nervous system, adrenal functioning, cholesterol production, Niacin(B3) is involved in lipid metabolism [62], amino acid and protein metabolism, and photosynthesis, Biotin is required in several specific carboxylation and decarboxylation reactions, Folic acid is required for normal blood cell formation and is involved as a coenzyme in one-carbon transfer mechanisms, blood glucose regulation, fish metabolism and improves cell membrane function and hatchability of eggs [63], Choline is essential for growth and good food conversion in fish[64], Inositol is a structural component in living tissues [65], Vitamin A is essential in maintaining epithelial cells, Vitamin E act as inter- and intracellular antioxidants to maintain homeostasis of labile metabolites in the cell and tissue plasma, Vitamin D functions as a precursor which stimulates the absorption of calcium from the intestine. [45] stated that 100 g dry matter of *Ulva sp* [66]. The contents of vitamins A (retinal), B1, B2, B3, B12, folic acid and C were 960 IU, 0.06, 0.03, 8.0, 6.3, 11.8 mg and 10.0 mg. Water-soluble vitamins such as vitamin C are present in large amounts in *Ulva lactuca* (sea lettuce) Phytonutrition [67] [68] and [69].

### Minerals

Seaweeds are known as an excellent source of minerals, especially sodium and iodine, due to their high polysaccharide content [46]. It contained a considerably high amount of minerals. Regarding the DRI, both kinds of seaweeds were notably rich in iodine apart from iodine, copper while *Ulva reticulata* was rich in potassium, manganese and ferrous [70]. Minerals are also necessary for the fish. Bones, teeth and scale tissues require lots of minerals [71]. The minerals also carry out many supportive functions. Actually in most cases the fish suffer from a lack of Calcium and Phosphorus and supplementing minerals will help to compensate for the deficiency but excess of some minerals also can be poisonous [72]. Therefore, mineral supplements should not be used without any information and just by random. Although fish can absorb some minerals from the water through their gills, receiving a supply of minerals in their diet is essential because of their involvement in skeleton formation, the regulation of the acid-base balance and osmotic and ionic regulation [47].

### **Carotenoids and colour**

Dietary carotenoids and carotenoid-protein complexes are the main source of fish skin and muscle pigmentation; therefore, to increase the skin and flesh colour in captivity, fish must obtain an optimum level of carotenoids in their diet. Fish skin colour is mainly attributed to the presence of chromatophores that contain pigments including melanins, pteridines, purines and carotenoids [73]. Since fish, like other animals, do not biosynthesize carotenoids de novo, it is essential to provide them with dietary supplements to allow the storage in tissue and teguments. Carotenoids are only synthesized by plants, phytoplankton (microalgae), zooplankton and crustaceans [74]. The most common carotenoids in freshwater include astaxanthin, zeaxanthin, xanthophylls, lutein,  $\alpha$ - and  $\beta$ -carotene, taraxanthin and tunaxanthin. The cost of synthetic pigments has encouraged the research of natural compounds such as green algae and even plant extracts as a pigment source. Therefore, owing to the carotenoid nutritional value is often lower than traditional diets, protein supplementation with this dietary source is highly recommended.

### **Fiber content**

Seaweeds have similar or slightly elevated levels of total fiber compared to terrestrial foodstuffs. *Ulva* spp. It contains total fiber 3.0 and soluble fiber 1.7 soluble fiber 1.3. The main components of this group are alginates, carrageenans, and agar, depending on the type of seaweed. These fibers are not digested to any great extent in the gut. Some show some fermentative capacity in the lower intestine, but the nature of soluble seaweed fibers is such that their passage through the gastrointestinal tract occurs largely without digestion [48].

### **Ash content**

[74] reported that the ash content 19.59% present in *Ulva* sp. *U. lactuca* has 24.6% dry weight, and *Ulva pertusa* 24.7% dry weight has ash content Behairy & El-[49] and [50].

### **Feed formulation**

Use of dietary algae as feed supplements may be expected to improve growth and digestive efficiency of feed [51]. *Ulva* sp. which contains the high amount of nutritional value and other potential effects of properties, less cost effect. So it will be highly useful in formation of feed and preparation. *Ulva reticulata* sp. has 35% protein, this will be used as basal diet for *Ulva* meal supplementation for fish health management (antimicrobial activity against fish pathogens), suggesting their potential for the development of anti-pathogenic agents, enhancing growth rate, and colouration [52], it will be a key role for producing quality ornamental fishes in India. [53] reported that 15% of *Ulva* sp was ideal to use as ingredients for feed formulation, this ratio of ingredients would be optimum diet for fishes, it also enhances the fish growth and survival. Similarly, *Ulva* sp. meal used as a feed supplementation, as a result, it increased growth rate and higher body protein gain concentration in common carp *Cyprinus carpio*. The fish however, is the subject to evaluate the nutritional adequacy of the feed in terms of increase in weight gain, higher reproductive capacity or reduced mortality and morbidity (CAN, 1993). Feed formulation is an art and its knowledge allows the formulator to take advantage of substituting one feedstuff for another and compound feed at the lowest cost, these studies supported to use as source of ingredients for fish feed formulation.

## **II. Conclusion**

Plenty of untapped seaweeds are available in our coastline, but the utilization is limited. So, by the utilization of the seaweeds from the coastal areas, the coastal people will get more employment opportunities. The *Ulva* sp. contains high levels of ash, appreciable protein and dietary fiber contents and relatively high levels of macro elements, essential amino acids and soluble and insoluble dietary fibers, and high iodine content. *Ulva*, a natural pigment source may enhance the potential of seaweed inclusion in fish feed and may perhaps replace and reduce artificial colourants currently used in the industry. Their nutritional compositions together with their physicochemical properties and positive effects to fish health supports our intention of recommending *Ulva* species as a potential feed to ornamental fishes. Using raw seaweed (containing rich protein, essential amino acids, lipids, vitamins, minerals, ash, carotenoids) as an ingredient of freshwater ornamental fish feed will improve the water quality which ensures environment safety.

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