Microalgae as a Potential Source of Bioactive Food Compounds

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Current Research in Nutrition and Food Science
www.foodandnutritionjournal.org
ISSN: 2347-467X, Vol. 09, No. (3) 2021, Pg. 917-927

Article History
Received: 25 September 2020
Accepted: 01 November 2021
Keywords
Therapeutic applications; Bioactive food Compounds; Microalgae; Nutraceuticals; Pigments.

Abstract
Microalgae are unicellular, photosynthetic organisms that can grow on diverse aquatic habitats like ponds, lakes, rivers, oceans, wastewater and humid soils. Recently, microalgae are gaining importance as renewable sources of biologically active food compounds such as polysaccharides, proteins, essential fatty acids, biopigments such as chlorophylls, carotenoids, astaxanthin, as well as vitamins and minerals. The bioactive food compounds of microalgae enable them to be part of multitude of applications in numerous industrial products for healthy life and ecosystem. This review article summarizes the applications of biologically active food compounds derived from microalgae as nutraceuticals, healthy dietary supplements, pharmaceuticals and cosmetics. Further, this review article highlights the importance of research focus on the identification and extraction of bioactive food compounds from the huge numbers of microlage that exist in nature for sustainable global food security and economy.

Introduction
Increasing awareness on the positive effect of diet on human wellbeing has brought novel natural ingredients and functional food products into a new extraordinary age.¹ The functional food is commonly defined as a diet comprising more than one useful

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Doi: http://dx.doi.org/10.12944/CRNFSJ.9.3.18
constituents that proffer supplementary health benefits in addition to the rudimentary, alimentary and energetic importance that each food offers. Functional foods have been linked with improved health and quality of life, good health promotion, and decreased threat of ailment. Public awareness on healthy diet is increasing in recent times as it is important for the prevention of chronic diseases including cancer, cardiovascular problems, and osteoporosis. Besides, the societal necessity to minimize the prescription of pharmaceutical products due to their adverse side effects and minimize the cost of healthcare, has encouraged the government agencies and the industries towards the wide usage of functional foods.

The utilization of microalga Nostoc to survive hunger among Chinese people has inferred that human beings had used microalgae as a food supplement over thousands of years. Certain blue-green microalgae such as, Aphanizomenon and Spirulina have been utilized for thousands of years by humans as food.

Microalgae comprise a vast diversity of microorganisms from prokaryotic cyanobacteria to eukaryotic microalgae that can synthesize bioactive substances by using carbon dioxide, nutrients (nitrogen, phosphorus, potassium) and solar energy proficiently. The carotenoids, long-chain fatty acids, sugars, both essential and non-essential amino acids, minerals, enzymes and vitamins are the most sought after bioactive compounds produced by the microalgae that are essential for human nutrition and wellbeing. Thus, microalgae are perceived as ideal candidates for modern "nutraceutical" or "pharma food".

Arthospira plantesis, Haematococcus pluvialis, Dunaliella salina, and Chlorella vulgaris are the most commonly used microalgae as food supplements in human and feed additives for animals as they are the most biotechnologically relevant microalgae. In addition, microalgae products are used in cosmetic industries as skin cream pigments and in pharmaceutical applications owing to their superior therapeutic values. Furthermore, microalgae function as probiotic agents and are incorporated in traditional food products such as pasta, biscuits, salad dressings and mayonnaise to enhance the intake of nutritionally diverse foods that promotes human health outcomes. Interestingly, microalgae are also used as a natural food colorant or as a dietary substitute in snack foods, pasta, gums, candy bars, snacks and drink mixes.

Due to the abundance of accessible microalgae bestowed in nature, selecting the most appropriate microalgae for specific applications in food technology is extremely important for successful production of novel foods. Conversely, biomass manufacturing systems that are currently in use have to be modified using more advanced technologies, in operational as well as technological levels, to enrich the bioactive compounds produced by the microalgae.

To highlight the importance of usage of the products synthesized and extracted from microalgae as an alternative source for naturally produced healthy food supplements, this review article summarizes the nutritional values and the therapeutic applications of the bioactive compounds derived from microalgae.

Bioactive Food Compounds of Microalgae
Microalgae are the fascinating life forms, whose evolution is not only to survive but also to flourish in the harshest surroundings of the planet. They do this by naturally generating a substantial collection of defensive and nourishing compounds like flavonoids, phospholipids, special carotenoids, antioxidants, oils rich in nutrients, non-digestible oligosaccharides and fatty acids. The biosynthesis of the bioactive compounds including natural antioxidants and drugs from microalgae are stimulated and enhanced by manipulating the cultivation procedures like increasing or decreasing the temperature, nutrients, growth phase, photoperiod and light intensity for optimal growth and high yield.

Lipids
The oils derived from microalgae can be a good substitute for the currently used vegetable oils as the microagal oils are rich in essential fatty acids. For an example, the concentrations of linoleic and alpha-linolenic acids in microagal oils are greater than the oils obtained from rape seed (canola), soy or sunflower oils.

The development of high concentrations of nutritionally important polyunsaturated long-chain fatty acids (PUFAs), such as docosahexaenoic
acid (DHA, 22:6, ω−3) and eicosapentaenoic acid (EPA, 20:5, ω−3) can be made possible through microalgae oils. The EPA and DHA supplements are well known for their beneficial effects to enhance brain function, especially to improve children's cognitive performance and also prevent cardiovascular diseases and inflammation. Interestingly, microalgae oils can be considered as a natural gift to vegetarians since the oils derived from microalgae can be a best substitute for fish oils. Also the chemical pollutants, like mercury, present in the fish supplies can contaminate the fish oils that in turn might cause health hazards to the consumers. Hence, microalgal oils can be a better alternative for fish oils available in the market.

Initially, the primary objective of the research development on algal lipids was targeted towards the synthesis of biodiesel. However, the wider usage of microalgal omega fatty acids in infant formulations and also as nutraceutical agents have attracted the current market considerably. Numerous microalgae strains have been analyzed for fatty acid content and great accumulations of DHA and EPA have been found in the strains of the genera *Nannochloropsis*, *Phaeodactylum*, and *Koliella*. Notably, the arachidonic acid of *Porphyridium cruentum* helps to improve normal growth, improve visual and functional development in infants. Similarly, the eicosapentaenoic acid present in *Nannochloropsis* sp. has beneficial effects on cardiovascular system as it offers protection against atherosclerosis and nervous sytem towards mental development and support.

**Proteins**

In the 1950s itself, microalgae were proposed as a novel protein source due to their high protein content and rich amino acid profile. The hydrolysates or peptides of microalgal proteins can be produced by various enzymatic processes and fermentation. The protein constituents of microalgae provide numerous health benefits such as antihypertensive, antioxidant, anticancer, immune-modulatory, anticoagulant, and hepato-protective. The microalgae commonly used in the formulation of protein products are *Spirulina* (65% protein), *Dunaliella* (57% protein) and *Chlorella* (55% protein). Special attention has been given to *Spirulina* among all the studied microalgae species due to its supreme quality and protein quantity (60–70 percent of dry weight) with easy digestability. Phycobiliproteins are a curious category of microalgae proteins, which are the accessory photosynthetic pigments, including phycocyanin, phycoerythrin, phycocerythrocyanin, and allophycocyanin. The *Synechococcus sp.* and *Arthrosepira sp.* are the most important algae currently used to extract phycobiliproteins. The phycobiliproteins are used in dairy products, chewing gums, sweets and ice creams as natural colorants and also find application in several nutraceutical products including tablets and capsules. Numerous beneficial health effects of phycobiliproteins including neuroprotective, antioxidant, hepatoprotective, anti-inflammatory, anticancer and hypocholesterolemic have been reported.

Solvent extraction, enzyme hydrolysis and microbial fermentation techniques are utilized to produce bioactive microalgae peptides. Enzymatic hydrolysis are preferred in food and pharmaceutical industries due to the absence of residual organic solvents or potentially harmful compounds in food. Advanced high throughput analysis and molecular studies of algal peptides are required to fully benefit the superior novel proteins from the microalgae.

**Carbohydrates**

A relatively low photo conversion efficiency of algae enables them to build up large carbon concentrations (greater than 50% by dry weight) with substantial biofuels in microalgae, which in particular act as protective, storage and structural molecules. The use of microalgae as a sustainable source of carbohydrates is a potential area that should be explored further. The glycogen (α1,4-based glucan), hybrid starch and amylopectin-like polysaccharides (starch) that are closely related to the plant are present in cyanobacteria, red algae and green algae respectively. Microalgae contain both prominent sugars like mannose, galactose, xylose, arabinose and glucose, and also less frequent sugars, including rhamnoses, fucose and uronic acids. Cultivation and environmental factors can modulate nearly 33-64% on the content of microalgae carbohydrates as they relate to the carbon source and metabolism (e.g. autotrophy, heterotrophy and mixotrophy).

Some microalgae polysaccharides can be used in industries commercially, taking into account...
of the rapid growth by crop control.\textsuperscript{31} Isolated polysaccharides from microalgae, such as \textit{P.cruentum}, \textit{S.platensis}, \textit{D. Salina}, \textit{Rhodella reticulate} and \textit{Schizochytrium sp.}, displayed useful antioxidant properties and efficient scavenging capabilities on superoxide radicals, hydroxyl radicals and hydroxyl peroxide.\textsuperscript{32} Recent research disclosed the occurrence of high content polysaccharides in \textit{Isochrysis galbana} (up to 25\% dry cell weight).\textsuperscript{33} The rich carbohydrate content of microalgae can serve as an alternative source of sustainable energy that can be supplemented to alleviate hunger and poverty.

**Phytochemicals**

Phytochemicals are group of bioactive compounds available in microalgae with diverse biological features including antioxidant, anti-inflammatory and antimicrobial activities. Phenolics are the most abundant phytochemicals that exist in microalgae and are well known for their antioxidant properties.\textsuperscript{34} The phenolic compounds are generally divided into 10 categories, namely phenolic acid, hydroxycinnamic acid, basic phenol, xanthone, flavonoid, stilbene acid, anthraquinone, coumarins, naphthoquinones and lignins.\textsuperscript{35} Flavonoids are the active scavengers for various forms of reactive oxygen species (ROS) and lipid peroxyl radicals to prevent lipid peroxidation and oxidative stress.\textsuperscript{34}

\textit{Arthrosira} extracts exhibited antioxidant activities by preventing the peroxidation of LDL cholesterol which leads to constraining atherosclerotic plaques and strokes.\textsuperscript{36} Antioxidant effects evaluated by testing oxidation stability demonstrated that chlorogenic and caffeic acids found in this microalgae have greater antioxidant activity than other phenolic acids.\textsuperscript{37} The combined effects of these phenolics and 13-cis-retinoic acid have been documented to not only prevent lipid peroxidation but also protective against various cancer.\textsuperscript{38} Numerous earlier findings have confirmed the antioxidant potential of phenolic compounds obtained from \textit{Chlorella} and \textit{Dunaliella}.\textsuperscript{37} The presence of these phytonutrients and bioactive substances make the microalgae a potent source of nutritional ingredients, nutraceuticals and dietary supplements.

**Vitamins and Minerals**

Microalgae also produce large amount of essential micronutrients such as vitamins and minerals.\textsuperscript{38} Microalgal biomass have been reported to constitute important vitamins (e.g., Thiamin (B1), Riboflavin (B2), Pyridoxine (B6), Cyanocobalamine (B12), vit C and vit E) and sufficient mineral content (e.g., sodium, potassium, copper, magnesium, iron and zinc). The cobalamin (vitamin B12) is commonly present in the green and red algae at high concentrations\textsuperscript{36} that makes microalgae as an alternative source of B12 especially for vegans and vegetarians.

Fabregas and Herrero performed a research in 1990 to find out the vitamin content of different strains of microalgae. They observed that, relative to traditional food sources, microalgae consist of a greater concentration of four vitamins; provitamin A, vitamin B1, vitamin E and folic acid. \textit{Dunaliella tertiolecta} has been confirmed to have the capacity to synthesize vitamin B12 (cobalamin), vitamin B2 (riboflavin), vitamin E (tocopherol) and provitamin A (β-carotene). \textit{Tetraselmis suecica} has also been an outstanding source of vitamin B1 (thiamin), vitamin B3 (nicotinic acid), vitamin B5 (pantothenic acid), vitamin B6 (pyridoxine) and vitamin C (ascorbic acid), and \textit{Chlorella sp} contains a high concentration of vitamin B7 (biotin). A research by Shim et al. found that about 9-18\% of \textit{Chlorella} species are a plentiful source of vitamin B12.\textsuperscript{39}

There are several bioactive food compounds derived from microalgae that can be employed in therapeutic applications. For instance, γ-Linolenic acid (GLA) and vitamins from \textit{S. platensis} are utilized as immunity boosters.\textsuperscript{40, 41} Proteins from \textit{S.platensis} and \textit{D.salina}, short chain fatty acids from \textit{H.pluvialis}, proteins and dietary fibers from \textit{Chlorella} and phenylethylamine from \textit{Aphanizomenon} are used as health food supplements.\textsuperscript{41,42,43,44,45} Besides, Eicosapentaenoic acid (EPA), Arachidonic acid (ARA) and vitamins derived from \textit{P.cruentum} are consumed as nutraceuticals to enhance the blood clotting and immune system.\textsuperscript{46, 47} Table 1 outlines the therapeutic applications of bioactive food compounds obtained from microalgae. These bioactive compounds are extensively used for various physiological functions and play a vital role in ensuring good health and well being.

**Pigments**

Presence of different pigments and colors in each phylum is one of the most noticeable characteristics
of microalgae. Given their phylogenetic age, it is self-evident that they have evolved to produce pigments that are unique to them. The different types of pigments isolated from microalgae have been proven to have numerous health benefits and hence have attracted the industries in producing various food and pharmaceuticals products in recent years using microagal pigments as an active food additive. Table 2 summarizes the pigments of microalgae and their potential applications. These pigments are natural with zero toxicity and better health outcomes that make them a healthier choice compared to the synthetic food additives and colorants.

**Table 1: Therapeutic applications of bioactive food compounds from microalgae**

<table>
<thead>
<tr>
<th>Microalga</th>
<th>Bioactive compounds</th>
<th>Physiological Functions</th>
<th>Therapeutic Applications</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Spirulina platensis</em></td>
<td>γ-Linolenic acid (GLA)</td>
<td>Maintains tissue integrity; delays aging</td>
<td>Immunity booster</td>
<td>40</td>
</tr>
<tr>
<td></td>
<td>Vitamin C, vitamin K, vitamin B12, vitamin A &amp; α-tocopherol Proteins</td>
<td>Antioxidant; forming blood cells; blood coagulation mechanism</td>
<td>Immunity booster</td>
<td>41</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Anticoagulant; immunomodulatory</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Dunaliella salina</em></td>
<td>Proteins</td>
<td>Antimicrobial; Antiproliferative</td>
<td>Health food supplement;</td>
<td>42</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>therapeutical</td>
<td></td>
</tr>
<tr>
<td><em>Haematococcus pluvialis</em></td>
<td>Short-chain fatty acids</td>
<td>Antimicrobial</td>
<td>Health food supplement</td>
<td>43</td>
</tr>
<tr>
<td><em>Chlorella vulgaris</em></td>
<td>Proteins</td>
<td>Anticoagulant; immunomodulatory</td>
<td>Health food supplement</td>
<td>44</td>
</tr>
<tr>
<td></td>
<td>dietary fiber</td>
<td>Detoxify toxic metals and pesticides</td>
<td>Health food supplement;</td>
<td>44</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>detoxifier</td>
<td></td>
</tr>
<tr>
<td><em>Porphyridium cruentum</em></td>
<td>Eicosapentaenoic acid (EPA)</td>
<td>Nutraceutical; antimicrobial; anti-inflammatory:</td>
<td>Baby and health food supplement</td>
<td>46</td>
</tr>
<tr>
<td></td>
<td>Arachidonic acid (ARA)</td>
<td>Platelets aggregation; vasoconstrictive</td>
<td>therapeutics; health ingredient</td>
<td>46</td>
</tr>
<tr>
<td></td>
<td>Vitamin C, vitamin K, vitamin B12, Vitamin A &amp; α-tocopherol Proteins</td>
<td>Antioxidant; blood cell formation; blood clotting mechanism</td>
<td>Immune system</td>
<td>47</td>
</tr>
<tr>
<td><em>Aphanizomenon</em></td>
<td>Phenylethylamine</td>
<td>Prevents neurodegenerative diseases</td>
<td>Healthfood supplement;</td>
<td>45</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>therapeutical</td>
<td></td>
</tr>
</tbody>
</table>

**Chlorophyll**

Chlorophylls are green in colour, which are non-polar pigments with a porphyrin chain and are present in cyanobacteria, algae, and superior plants. Chlorophylls contain tetapyrrole with tightly bound magnesium atom. Chlorophylls are
Monomeric pigments in green algae (Chlorophyta). Microalgae contain 0.5 to 1.0% of chlorophyll per gram. Chlorophyllin is a chlorophyll derivative in which sodium or copper replaces magnesium and the phytol chains are lost. Chlorophyllins are used in dietary supplements to control geriatric patients' body odor and studies have reported the antimutagenic and anticarcinogenic effects of chlorophyll and chlorophyllin.

Table 2: Microalgal pigments and their potential applications

<table>
<thead>
<tr>
<th>Microalgae</th>
<th>Pigment</th>
<th>pigment Colour</th>
<th>Applications</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Spirulina platensis</em></td>
<td>Chlorophyll</td>
<td>Green</td>
<td>Pharmaceutical and cosmetics</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>Phycocyanin</td>
<td>Blue-green</td>
<td>Dyes, beverages, whipped cream, hot cream and sweets, cosmetics, antiviral</td>
<td>61, 62</td>
</tr>
<tr>
<td><em>Dunaliella salina</em></td>
<td>β-Carotene</td>
<td>Yellow</td>
<td>Pro-vitamin A, an antioxidant food, additive E160a; egg yolk colorant</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td>Bixin</td>
<td>Yellowish to peach color</td>
<td>Food additive E160b (colorant); cosmetics</td>
<td>10, 61</td>
</tr>
<tr>
<td></td>
<td>Lutein</td>
<td>Yellow-orange</td>
<td>Dietary additive E161b, gg yolk coloring, animal tissue pigmentation, medicinal (anti-macular degeneration), cosmetics (coloring)</td>
<td>64</td>
</tr>
<tr>
<td><em>Haematococcus pluvialis</em></td>
<td>Astaxanthin</td>
<td>Reddish-salmon</td>
<td>Livestock additive E161j, antioxidant, salmon and trout farming (color, immune response)</td>
<td>65</td>
</tr>
<tr>
<td></td>
<td>Canthaxanthin</td>
<td>Golden-orange</td>
<td>Food additive E161 g, salmonid farming; tanning pills</td>
<td>66</td>
</tr>
<tr>
<td><em>Chlorella vulgaris</em></td>
<td>(α-Tocopherol)</td>
<td>Brown</td>
<td>Vitamin E, food additive E306, E307, E308. Antioxidant in cosmetics and foods,</td>
<td>63</td>
</tr>
<tr>
<td></td>
<td>Chlorophyll</td>
<td>Green</td>
<td>Pharmaceutical cosmetics</td>
<td>10</td>
</tr>
<tr>
<td><em>Porphyridium cruentum</em></td>
<td>Phycoerythrin</td>
<td>Red</td>
<td>Antiviral</td>
<td>67</td>
</tr>
<tr>
<td><em>Aphanizomenon</em></td>
<td>Phycocyanin</td>
<td>Blue-green</td>
<td>Food dyes (beverages, ice cream, sweets), cosmetics, histochemical fluorescent markers, antibody marks against receptors and other biological molecules</td>
<td>61</td>
</tr>
</tbody>
</table>

**Carotenoids**

Among the lipid components of the microalgae, some lipophilic components are especially important in health industry. Because of easy growing, non-competitive food production processes, and adaptation to the changing environmental conditions.
which in turn results in the production of wide range of secondary metabolites, microalgae have recently generated a high level of interest for different natural sources of carotenoids.\textsuperscript{51} Carotenoid biosynthesis can be induced by regulation of crop circumstances or genetic engineering approaches.\textsuperscript{52} In microalgae, carotenoids appear to be mainly photo-protective and light-processing pigments. Carotenoids have high potential antioxidant properties, and hence protect the harmful effects of excessive UV solar radiation and oxidative stress by scavenging free radicals.\textsuperscript{10, 53}

Several groups of carotenoids are found to be effective against more than 60 diseases that are life-threatening including cancers, cardiovascular problems, premature aging, and arthritis.\textsuperscript{54} The microalgae \textit{H. pluvialis} is the primary source of carotenoid (β-carotene). \textit{Dunaliella} extract has been sold as dietary supplements in numerous places since 1980.\textsuperscript{55} The highest level of 9-cis-beta-carotene is found in \textit{Dunaliella} amongst all the natural sources. Healthcare and consumer industries prefer natural beta-carotenones from microalgae as they are better absorbed in living organisms by a combination of trans and cis isomers than the beta carotenones synthesized by chemical processes.\textsuperscript{56} Fortification of beta-carotene in soft drinks, cheese, butter and margarine is becoming popular in recent times. In addition, most cyanobacteria are α-carotene generators.\textsuperscript{56}

The carotenoids are proven to have anticancer actions through activation of cell apoptosis and suppression of cell proliferation. In particular, beta-carotenones such as astaxanthin, canthaxanthin, and zeaxanthin help to reduce the size and number of liver neoplasms.\textsuperscript{57} Astaxanthin has benefited humans through improved eye protection, strength and endurance, and also by preventing premature aging, inflammation and UV-A damages.\textsuperscript{58} Several positive effects of astaxanthin on vision, growth, immune function, reproduction, and regeneration have brought in the usage of this pigment in human nutrition and animal feed.\textsuperscript{58} Astaxanthin was approved in 1987 as a food preservative for utilization in aquaculture.\textsuperscript{59}

\textit{H. pluvialis} is a green freshwater microalgae that can produce significant amounts of astaxanthin under oxidative stress. Currently, several companies grow algae on a wide scale using different methods to produce cysts with enriched astaxanthin. Using various methods of extraction, the \textit{H. pluvialis} can yield about 70–94% of astaxanthin which is a boon to promote blue economy.\textsuperscript{60}

Natural pigments isolated from microalgae have been proven to have numerous health benefits and have been utilized in many industrial products. Chlorophyll and phycocyanin from \textit{S. platensis}, α-tocopherol and chlorophyll from \textit{C. vulgaris} as well as phycocyanin from \textit{Aphanizomenon} are vastly applied in food, cosmetics and pharmaceutical sectors.\textsuperscript{10, 61, 62, 63} Besides, \textit{D. salina} contains pigments like β-carotene, bixin, and lutein. They are mostly yellow to orange in colour and are used in food additives and cosmetics, and also for animal tissue pigmentation.\textsuperscript{10, 61, 64} Other than that, the reddish and golden orange pigments of \textit{H. pluvialis} such as astaxanthin and canthaxanthin are widely as livestock additive for salmon and trout farming to produce coloured fish with enhanced antioxidant and immune properties.\textsuperscript{65, 66} Further, the phycoerythrin derived from \textit{P. cruentum} has proven to have antiviral properties to be utilized as a nutraceutical agent.\textsuperscript{67}

**Conclusion**

Currently the industrialized microalgae are primarily used as food, food additives, aquaculture feed, dyes, cosmetics, pharmaceuticals, and nutraceuticals. For human usage, only a small fraction of the total number of algal organisms are cultivated. There exist possibly numerous species of microalgae that nature has bestowed with good nutritional values and health benefits which remain under explored for their potential usage. Therefore, in the forth coming years, the potential of microalgal use in food intake, nutritional supplements, energy production, and much more is likely to intensify. Geographical use of microalgae for human nutrition will offer economically manageable and naturally produced healthy food for sustainable supply of food to meet the demand of growing population of the world.

**Funding**

The author(s) received no financial support for the research, authorship, and/or publication of this article.

**Conflict of Interest**

The authors declare that they have no conflicts of interest.
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