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Microalgae Derived Alkenones and their Applications

Kyle McIntosh[1], David J Terrero[2], Rahul Khupse[3] and Amit K Tiwari[1]*

[1]*Department of Pharmacology and Experimental Therapeutics, University of Toledo, USA*

[2]*Pontificia Universidad Católica Madre y Maestra, Dominican Republic*

[3]*College of Pharmacy, University of Findlay, Findlay, USA*

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Corresponding author: Amit K Tiwari, Department of Pharmacology and Experimental Therapeutics, College of Pharmacy & Pharmaceutical Sciences, University of Toledo, Ohio 43614, Email: amit.tiwari@utoledo.edu; Tel:+1-419-383-1913; Fax:+1-419-383-1909

Abstract

Marine microalgae and their extracts, in recent years, have found applications, including use as biofuels, medicines and food. Recently, alkenones, a unique chain of long-chain lipids, extracted from Isochrysis microalgae, has emerged as a useful by product. This is the first review that summarizes the potential applications and limitations of alkenones use in paleoclimatology, biofuel, personal care industry and therapeutics.

Keywords: Alkenones; Microalgae; Paleoclimatology; Biodiesel; Personal care products

Abbreviations: CO_2: Carbon Dioxide; HPLC: High-Performance Liquid Chromatography; MAAs: Mycosporine-like Amino Acids; EPA: Eicosapentaenoic Acid; DHA: Docosahexaenoic Acid

Introduction

Marine algae have many advantages when compared to traditional terrestrial plant agriculture. One significant advantage is the lack of soil for growth; thus, algae do not compete with agricultural products for a limited growth area [1]. Furthermore, marine algae consume less water compared to land crops and yield a high production per acre due to their high growth rate. For these reasons, culturing algae, compared to terrestrial plants, should be cost-effective. Microalgae species or plankton are the primary producer of organic matter in aquatic environments by photosynthesis and provide a food source for the aquatic habitat. Microalgae can use CO_2, nutrient salts, organic matter, trace elements and they convert energy from the sun by photosynthesis [2].

Microalgae can be found in either freshwater or seawater environments. When exposed to extreme conditions, marine microalgae must adapt to new environments [3]. They typically produce diverse, secondary biologically active metabolites that are not present in terrestrial plants [4]. There are an estimated 200,000 to several million species of microalgae [5], and only 40,000 have been identified [2], and their unique metabolites could be of importance in the development of cosmetic products. Currently, marine microalgae derived from harvested seaweed is used as animal feed, wastewater treatment, carbon dioxide sequestration, biofuels, pharmaceuticals and nutraceutical and cosmetics applications [6].

Compounds obtained from Marine Microalgae

Several extraction and analytical techniques have been used in the past to extract beneficial compounds from marine microalgae. The Soxhlet extraction has been used for nonpolar, lipid derivatives, while High-Performance Liquid Chromatography (HPLC) can identify carbohydrates, carotenoids, fatty acids and mycosporine-like amino acids from marine algae [7]. A recent review [8] discussed additional extraction techniques, including supercritical-fluid extraction, enzyme-assisted extraction and microwave-assisted extraction. Several compounds of interest were extracted from algae, including mycosporine-like amino acids (MAAs) [9], glucosyl glycerols, polysaccharides/sulfated polysaccharides [10], polyphenols, and lipids [11] among others. Many of these compounds have been reported to have anti-inflammatory, antioxidant, and photo protection [12], anti-diabetic, anti-microbial, anti-cancer [13,14], gastroprotective and neuroprotective efficacy. The focus of this mini-review paper will be on the alkenones derived from Isochrysis galbana.

What are Alkenones?

Currently, only four strains of haptophytes from the order of Isochrysidales: *Emiliania huxleyi*, *Gephryocapsa oceanica*, *Isochrysis galbana*, and *Chrysotila lamellose*, are known to produce compounds referred to as either alkenes or alkenones [15]. Both

Chrysotila lamellose and *Isochrysis galbana* can produce C_{31}, C_{33}, C_{37}, and C_{38} alkenes [16], as well as C_{37}-C_{39} alkenones [17]. The difference between alkenes and alkenones is in the unsaturated methyl or ethyl ketone group [18]. The *Isochrysis sp.* is a non-toxic food source for cultivated marine animals [19] as it produces essential polyunsaturated fatty acids, such as Eicosapentaenoic acid and docosahexaenoic acid for the growth and development of fish larvae [20]. This marine alga is characterized by a lack of toxins, a quick growth rate and salinity tolerance [21]. Also, *Isochrysis sp.* lack cell walls, allowing for easier lipid extraction compared to other algae [22]. The most common cultured version is the Tahitian strain, "T-iso" [23].

Alkenones are identified by a methyl or ethyl ketone group, two to four non-methylene trans-double bonds and by long hydrocarbon chains (36-40 carbons)[24]. These alkenones were discovered to have a high melting temperature (71.1-77.4 °C),[25] and were compatible with other commonly used waxes in lipsticks, lip balms, and creams, thereby making them a potential wax-like compound for personal care products. Figure 1 shows the three major structures of alkenones isolated from *Isochrysis galbana.* The relative percentage of 0.1 grams of mixed alkenones derived from purified *Isochrysis* biomass was reported as 26.1% (37:3 methyl alkenones), 62.7% (37:2 methyl alkenones), and 15.7% (38:2 ethyl alkenones) (Figure 1) [26].

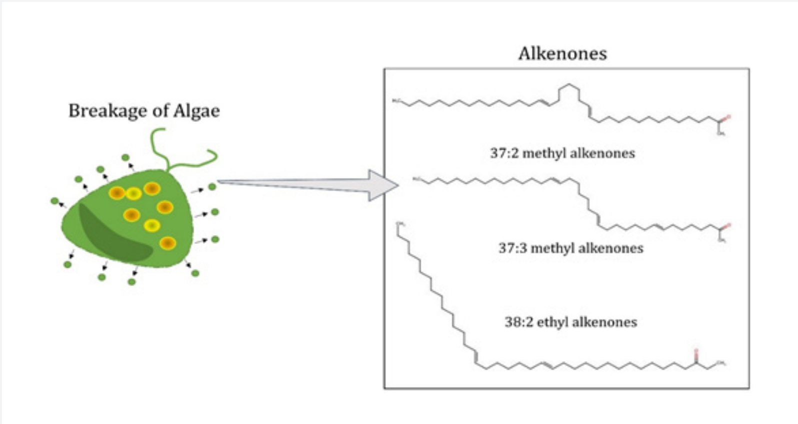

Figure1: Three common structures of alkenones derived from *Isochrysis galbana* when isolated for biofuel. The relative percentage from 0.1 gram of mixed alkenones was previously reported as 26.1% (37:3 methyl alkenones), 62.7% (37:2 methyl alkenones), and 15.7% (38:2 ethyl alkenones) (26).

Current Uses of Alkenones

Paleoclimatology

It is essential to accurately monitor climate change-driven trends that occur in the ocean. By analyzing the preserved alkenones and other lipids in the ocean bottom sediments, the alkenones structure can reflect the water temperature in that particular zone [26]. The number of unsaturated bonds in an alkenone is known to increase when the microalgae are grown or maintained at low temperatures [27]. This characteristic, an unsaturated index formula, termed $U^{k'}_{37,}$ is calculated as a parameter of the ratio of $C_{37:2}$ to $C_{37:3}$ alkenones [28,29]. The significant correlation between alkenones abundances and sea surface temperatures make the alkenones a widely used biomarker for paleoclimate studies. It should be noted that a common misconception is that distinct alkenones signature alone can directly identify specific species of microalgae. This theory was disproved after sampling 15 alkenone-containing lake surfaces indicated that DNA fingerprinting is a better approach to identifying species [30].

Biofuel

Isochrysis sp. is commercially cultivated algae grown worldwide [31], and having an algae strain that can yield high lipid production is a desirable characteristic for biodiesel production. Biodiesel derived from algae was shown to be less toxic in seawater when compared to conventional jet and marine diesel fuels [32]. In addition to having lower amounts of sulfur and particulate matter emissions, biodiesel increases performance in brake thermal efficiency (ratio of brake power to the thermal energy input from the fuel) [33]. Biodiesel also improved brake - specific energy consumption (a measure of the combustion quality of the fuels) performance for a single-cylinder, 4-stroke, air-cooled, direct injection diesel engine.

Lipids are an excellent source for making biofuel as they store high amounts of energy. *Isochrysis galbana* has an oil content up to 25 – 33 % dry weight [34]. For a discussion of lipid extraction technologies, see [35] A six-step protocol has been published that provides details about isolating and purifying alkenones from commercially available *Isochrysis* paste. Alone, alkenones with long chains of 37-39 carbons are not suitable for use jet fuel, but once converted to 8-13 carbons in length, they can be used for jet fuel [36]. Alkenones also have competitive thermal properties similar to other commonly used petroleum-derived phase change materials (PCMs) [37].

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Personal Care Products

The chemical and physical properties of alkenones that were derived from *Isochrysis sp*. physical have been characterized in terms of melting point, thickening capability of emulsions, viscosity and stability in personal care products. These results lead to alkenones being investigated in sunscreens combined with three "reef safe" UV filters [37]. The results from that study indicated that alkenones significantly increased the *in vitro* sun protection factor of the three "reef-safe" UV filters. Currently, there have only been limited studies on alkenones from *Isochrysis galbana* for medical applications. In a recent pilot study in female Sprague Dawley rats with these alkenones, we found that alkenones were safe following oral and dermal application (unpublic shed findings). Other studies have focus on two vital fatty acids, Eicosapentaenoic acid (EPA) [38-40] and docosahexaenoic acid (DHA) [41-44], which can affect blood pressure, lipoprotein metabolism and platelet and endothelial function. It is possible that alkenones derived from *Isochrysis* could potentially produce similar effects.

Conclusion

The many valuable compounds that can be derived from marine algae are the reason why agricultural farms are increasing as consumers are turning towards greener products. Alkenones extracted from Isochrysis sp. represent a major category of valuable compounds. Its usage was limited to a powerful paleo-oceanographic tool but has now expanded to biodiesel fuel and personal care products. It will be interesting to see what other fields of research these alkenones will play a role and how they will benefit our way of life.

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