



Microalgal Carotenoids and their applications

Pritikrishna Majhi, SudiptaMaity, Samarpita Dash, Anwesa Jena, Manoja Dash, DBT-BIRAC,
EYC,GIETU, Gunupur, Odisha

SouravSahil, Chintu K. Sah, AakashKumbhar, K. Kali Prasad Dora, ManjushDevnath,Department of
Biotechnology, GIETU, Gunupur, Odisha

Corresponding author mail address: pkmajhi94@gmail.com

Abstract- Nutraceuticals have gained growing attention in regards to their scientific investigation during the last few decades because of the growing demands. Therefore, the commercial microalgal cultivation for human nutritional products in and around the world is being emphasized currently in more or less. Microalgae synthesize varieties of natural products such as proteins, lipids, polysaccharides, enzymes, vitamins, minerals and pigments like phycocyanin, carotenoids and chlorophylls. Three most well-known genera (*Chlorella*, *Spirulina*, and *Dunaliella*) represent around 85% of the worldwide microalgal based compound production. Among all the metabolites from microalgae, pigments are the most significant ones in nature due to their higher industrial potential. Carotenoids are one of the metabolite among them. They are highly essential for normal vision and treatment of several degenerative diseases. There are approximately more than 400 carotenoids found in the nature. This review article compiled the importance of microalgal carotenoids, their resources, factors affecting their production and their global market strategy.

Keywords- Microalgae, carotenoids, applications, factors, global market

I. INTRODUCTION

Currently, microalgae have been attracted towards intensive research interests because of their high economical and commercial potential [1]. The varieties of bioproducts from microalgae can be extensively used in different nutraceuticals, pharmaceuticals, food colorants, additive and animal feed industries [2]. In comparison to the higher plants, fungi, and bacteria, microalgae employ a series of benefits involving easy availability, faster growth, shorter doubling and period higher yield. Hence, various phylum of microalgae (Chlorophyte, Cyanophyta, Erythrophyta and Chrysophyta) of great economic values are now being cultivated in bulky amounts [3, 4, 5] to get the desired high value product. Therefore, they are broadly used in the field of industrial manufacture of many bioactive compounds [6]. It has been also observed that thousands of bioactive compounds from eukaryotic microalgae and

more than 200 bioactive compounds can be extracted from blue green algae [6]. Among the different bioactive components, carotenoids are considered as one of the most significant high valued commercialized bioactive compound. They can synthesize carotene, lutein, violaxanthin, neoxanthin, astaxanthin, and fucoxanthin in large quantities, and are recognized as provitamin A [7]. It is one of the most important and abundant dietary Provitamin A supplement for human health which can be then converted into vitamin A i.e. retinol [8]. This carotene pigment is a ubiquitous compound related to the photosynthetic process in microalgae, having several kinds of biological activities [9]. Its raw extract and pure compound form possess some kind of bioactivities like hepato-protection, anti-obesity, anti-cancer, anti-inflammation, immune-modulation and [10, 11, 12]. Microalgal strains such as *Spirulina*,



Chlorella, *Dunaliella*, and *Haematococcus* are the major source of carotenoids [13].

Carotenoid is a redish-orange pigmented compound having tetraterpene with eleven conjugated double bonds [14]. Basically, it acts as an antioxidant by protecting active cells from oxidation reactions and also as an accessory pigment [15, 16]. This can be used as a food, food colorant, feed additive and precursor for the synthesis of aromatic antioxidants [17]. The chlorophyta strains *Dunaliella salina* and *Haematococcus pluvialis* are reported to accumulate the highest amounts of astaxanthin and β -carotene (up to 7% and 13% on dry weight basis, respectively) and hence, are assumed to be sustainable feedstock for the commercial production of carotenoids [18]. Thus, from an economical and feasibility point of view, it becomes advisable to optimize all the growth factors for recovery of carotenoids from microalgal cells. According to Wolf *et al.*, 2021, the global market value of β -carotene in the year of 2015 was \$432.2 million, while algae based β -carotene value was \$164.2 million [15, 19]. Hence, this research article summarizes the application, factors affecting carotenoid production, biosynthetic pathway and market growth of microalgal carotenoids in brief.

II. APPLICATIONS OF CAROTENOIDS

Astaxanthin and β -carotene are the two most important carotenoid compounds while lutein, zeaxanthin and lycopene are having less importance [20, 2, 12]. The major applications of carotenoid pigments are enlisted below:

- Natural food coloring agent in the form of pro vitamin A
- Additive to cosmetics (Sunscreen formulation)
- Anti-angiogenic and cardio protective activity

- Antioxidants preventing oxidative reactions
- Additive for animal feed,
- Additive for therapeutic, and nutraceutical applications
- They possess native anti-inflammatory properties due to their quenching action on ROS
- They also act as therapeutic chemo-preventive anti-cancer agent
- They can be used as antiaging anti anti-hypersensitive agent
- Application in agriculture
- They can be employed for wastewater treatment
- They are the substitute of fatty acids for biofuel production

III. FACTORS AFFECTING CAROTENOID SYNTHESIS

There are several growth factors such as nutrient media, pH, salinity, temperature, light intensity, photoperiod and composition which affect the growth of the desired organism [21]. Several stress conditions enable the strain to adapt the environment by releasing some sorts of bioactive components and in this way the production quantity of the bioactive molecule may increase or decrease. Nutrients required for microalgae are mainly of two categories such as macronutrients and micronutrients/ trace metals [22]. Differences in the culture media composition in more or less amount affect the growth of the strain which is inversely proportional to the carotenoid production. Another study by Bonnefond *et al.*, 2017, it has been observed that beta-carotene amount is strongly anti-



correlated with nitrogen quota emphasizing its carbon sink activity in nitrogen deprived conditions [23]. Similarly, pH of the culture medium is one of the significant environmental factors that affect the microalgal growth. The major fact which is essential for maintaining the initial pH of culture medium and the synthesized compound by the strain in the same culture medium during the growth phase. Moreover, the culture growth and composition of bioactive molecules produced by many algal species are highly affected by temperature other than its optimal value. It may stimulate or inhibit various intracellular activities by changing the functionality of numerous intracellular enzymes. Salinity is another essential factor that affects the growth of biomass and the quantity and quality of extracted beta-carotene. It has been observed that increased salinity has extensively reduced the cell growth while increase the amount of beta-carotene production. In addition to that light intensity has an important influence on the cellular metabolism of photosynthetic microbes. It plays key role in the bioavailability of various pigments in microalgal cells [20]. Hence, it is always advisable to maintain the culture of the specific algal strain as per its optimized or controlled conditions in order to attain highest amount of the desired high value product.

IV. MAJOR ALGAL STRAINS CONTAINING VARIOUS FORMS OF CAROTENOIDS

Carotenoid pigment is available in several forms and the significant carotenoid compounds are β -carotene, lutein, astaxanthin, zeaxanthin, lycopene and canaxanthene. Table 1. shows the major algal strains producing those essential pigments after overcoming their carotogenesis phase of growth.

TABLE I

MAJOR CAROTENOIDS AND THEIR MICROALGAL SOURCE

Name of the pigment	Name of the algal strains	References
01 β -carotene	<i>Chlorella</i> sp., <i>Dunaliella</i> sp., and <i>Haematococcus</i> sp., <i>Heterochlorella luteoviridis</i> , <i>Coelastrasteriastriolata</i> var. <i>multistriata</i>	[22, 24, 25]
02 Lutein	<i>Chlorella protothecoides</i> , <i>Dunaliella salina</i> , <i>Scenedesmus almeriensis</i> , <i>Parachlorella</i> sp. JD-076	[2, 25]
03 Astaxanthin	<i>Haematococcus pluvialis</i> , <i>Chlorella zofingiensis</i> , <i>Coelastrasteriastriolata</i> var. <i>multistriata</i>	[2, 25]
04 Zeaxanthin	<i>Chlorella ellipsoidea</i> , <i>Dunaliella salina</i> , <i>porphyridium purpureum</i>	[2, 20]
05 Lycopene	<i>Chlorella marina</i>	[2]
06 Canaxanthene	<i>Chlorella emersonii</i> , <i>Chlorella zofingiensis</i> , <i>Scenedesmus komarekii</i> , <i>Coelastrasteriastriolata</i> var. <i>multistriata</i>	[22, 25]

V. BIOSYNTHESIS OF CAROTENOID IN MICROALGAE

Microalgal Carotenoids are categorized as primary and secondary ones as per their accumulation property such as photosynthesis and under adverse conditions, respectively [26]. Initial phase of carotenoid synthesis starts in the chloroplast through the methylerythritol phosphate pathway (MEP) where 3X isopentenyl pyrophosphate (IPP, C5) and its isomer 1X dimethylallyl diphosphate (DMAPP, C5) are combined at a ratio of 3:1 in presence of geranyl diphosphate synthase (GPPS) and geranylgeranyl pyrophosphate synthase (GGPPS) to produce geranylgeranyl pyrophosphate (GGPP, C20). Fig.1.

summarizes the carotenoid synthesis pathway. Those biosynthesized Carotenoids can be deposited inside or outside of the chloroplast [2].

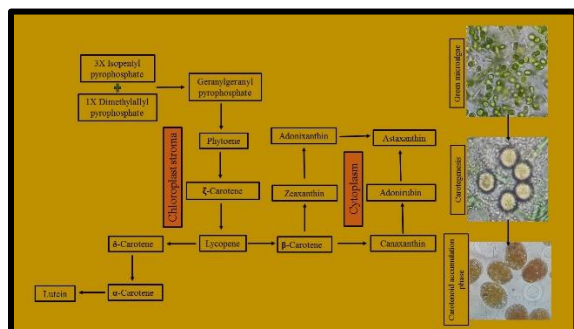
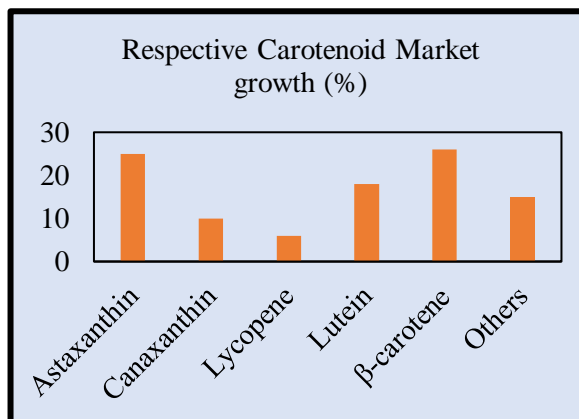


Fig.1. Biosynthetic Pathway of carotenoid production from microalgae

VI. MARKET POTENTIAL OF CAROTENOIDS IN THE PAST FEW YEARS

The international carotenoid market has expanded its growth up to 1.3 billion euros in the year of 2017 while it was predicted to be 1.8 billion euros by 2022 along with a CAGR of 5.7% in the subsequent 5 years [27, 28]. As per the Market Data Forecast report (2023-2028), it has been estimated that the global algae based carotenoid market was US\$1.57billion in 2022 and will more like reach US\$2.09billion by the end of 2028 with CAGR of 4.5% [29]. Similarly, according to the Grand View Research, (2015) data report, the global market value of β -carotene in the year of 2015 was US\$432.2 million, while algae based β -carotene value was \$164.2 million [19]. Moreover, the Deinove biotechnology company has also reported that 1400 metric tons of carotenoids were produced in the year of 2014 accounting for US\$1.4 billion. Another market research by Custom Market Insights has been observed that the demand of Global Carotenoids Market size & share revenue was approximately US\$1.5 Billion in 2021 which was expected US\$1.6 billion in 2022 and is expected to reach near about US\$2.1 Billion by

2030, at a CAGR of 3.5 in between 2022 to 2030. Moreover, the total commercial value of whole algal biomass production in each year is estimated about 10,000 tons. Fig.2 shows the respective market growth of some carotenoid compounds extracted from



Haematococcus sp. and *Dunaliella* sp. during the year of 2019.

Fig. 02. The respective market growth of some carotenoid compounds

VII. FUTURE PROSPECTS AND CONCLUSION

The future prospects in this regard should involve microalgal genetic modifications for salt tolerance and high Carotenoid accumulation property. Many microalgae, such as *Dunaliella salina*, *Scenedesmus* sp., *Haematococcus pluvialis*, and *Chlorella* sp., are considered as foremost economic and sustainable sources of carotenoids as well as other bioactive molecules. There are few studies aiming at making feasible the large-scale production of microalgae by introducing various types of cultivation systems and operational growth factors. Although microalgae possess a great diversity still major of the part of this organism has been remained unexplored except the few aforementioned strains. Hence, extensive study must be done by taking those unexplored part of numerous novel microalgal strains



proving them different extreme environment to grow and excrete high value bioactive components. Various indigenous strains should be collected, identified and exploited to explore their bioactive component content and then should be commercialized in order to increase its economic value as well as to develop its geographical habitat following adaptive evolution.

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REFERENCE

1. Sanghvi, A.M.; Lo, Y.M. Present and potential industrial applications of macro- and microalgae. *Recent Pat. Food Nutr. Agric.*, 2, 187–194, 2010.
2. Cezare-Gomes EA, Mejia-da-Silva LD, Pérez-Mora LS, Matsudo MC, Ferreira-Camargo LS, Singh AK, de Carvalho JC. Potential of microalgae carotenoids for industrial application. *Applied biochemistry and biotechnology*, 15, 188:602-34. 2019.
3. Kratzer, R.; Murkovic, M. Food Ingredients and Nutraceuticals from Microalgae: Main Product Classes and Biotechnological Production. *Foods*, 10, 1626, 2021.
4. Vieira, M.V.; Pastrana, L.M.; Fuciños, P. Microalgae Encapsulation Systems for Food, Pharmaceutical and Cosmetics Applications. *Mar. Drugs*, 18, 644, 2020.
5. Ambati, R.R.; Gogisetty, D.; Aswathanarayana, R.G.; Ravi, S.; Bikkina, P.N.; Bo, L.; Yuepeng, S. Industrial potential of carotenoid pigments from microalgae: Current trends and future prospects. *Crit. Rev. Food Sci. Nutr.*, 59, 1880–1902, 2019.
6. Barkia, I.; Saari, N.; Manning, S.R. Microalgae for High-Value Products Towards Human Health and Nutrition. *Mar. Drugs*, 17, 304, 2019.
7. Foong, L.C.; Loh, C.W.L.; Ng, H.S.; Lan, J.C. Recent development in the production strategies of microbial carotenoids. *World J. Microbiol. Biotechnol.*, 37, 12, 2021.
8. Tanguy, G.; Legat, A.; Gonçalves, O.; Marchal, L.; Schoefs, B. Selection of Culture Conditions and Cell Morphology for Biocompatible Extraction of β -Carotene from *Dunaliella salina*. *Mar. Drugs*, 19, 648, 2021.
9. Chen, W.; Chen, G. The Roles of Vitamin A in the Regulation of Carbohydrate, Lipid, and Protein Metabolism. *J. Clin. Med.*, 3, 453–479, 2014.
10. Marcelino, G.; Machate, D.J.; Freitas, K.d.C.; Hiane, P.A.; Maldonado, I.R.; Pott, A.; Asato, M.A.; Candido, C.J.; Guimarães, R.d.C.A. β -Carotene: Preventive Role for Type 2 Diabetes Mellitus and Obesity: A Review. *Molecules*, 25, 5803, 2020.
11. Riccio, G.; Lauritano, C. Microalgae with Immunomodulatory Activities. *Mar. Drugs*, 18, 2, 2019.
12. Martínez Andrade, K.A.; Lauritano, C.; Romano, G.; Ianora, A. Marine Microalgae with Anti-Cancer Properties. *Mar. Drugs*, 16, 165, 2018.
13. Wang J, Hu X, Chen J, Wang T, Huang X, Chen G. The extraction of β -carotene from microalgae for testing their health benefits. *Foods*. 10:11(4):502, 2022.
14. Cohen, G. N., Biosynthesis of Carotene, Vitamin A, Sterols, Ubiquinones and Menaquinones, in *Microbial Biochemistry*,



- Springer Netherlands, Dordrecht, 523–538, 2014.
15. Sathasivam R, Ki JS. A review of the biological activities of microalgal carotenoids and their potential use in healthcare and cosmetic industries. *Marine drugs*, 12:16(1):26, 2018.
 16. Wolf L, Cummings T, Müller K, Reppke M, Volkmar M, Weuster-Botz D. Production of β -carotene with *Dunaliella salina* CCAP19/18 at physically simulated outdoor conditions. *Engineering in Life Sciences*, 21(3-4):115-25, 2021.
 17. Woortman, D. V., Jürgens, S., Untergehrer, M., Rechenberger, J. *et al.*, Greener aromatic antioxidants for aviation and beyond. *Sustainable Energy Fuels*, 4, 2153–2163, 2020.
 18. Rammuni MN, Ariyadasa TU, Nimarshana PH, Attalage RA. Comparative assessment on the extraction of carotenoids from microalgal sources: Astaxanthin from *H. pluvialis* and β -carotene from *D. salina*. *Food chemistry*. 277:128-34, 2019.
 19. Grand View Research, Beta-Carotene Market Analysis By Source (Algae, Fruits & Vegetables, & Synthetic), By Application (Food & Beverages, Dietary Supplements, Cosmetics, & Animal Feed), 2015
 20. Rajesh K, Rohit MV, Mohan SV. Microalgae-based carotenoids production. *In Algal green chemistry*, 139-147, 2017.
 21. Majhi P, Samantaray SM. Thermotolerant microalgal diversity in the chromium metal polluted sites of Sukinda mining area. *International Journal of Current Microbiology and Applied Sciences*, 9(3):1109-20, 2020.
 22. Pourkarimi S, Hallajisani A, Alizadehdakhl A, Nouralishahi A, Golzary A. Factors affecting production of beta-carotene from *Dunaliella salina* microalgae. *Biocatalysis and Agricultural Biotechnology*, 29:101771, 2020.
 23. Bonnefond H, Moelants N, Talec A, Mayzaud P, Bernard O, Sciandra A. Coupling and uncoupling of triglyceride and beta-carotene production by *Dunaliella salina* under nitrogen limitation and starvation. *Biotechnology for Biofuels*, 10:1-0, 2017.
 24. Jaeschke DP, Menegol T, Rech R, Mercali GD, Marczak LD. Carotenoid and lipid extraction from *Heterochlorellaluteoviridis* using moderate electric field and ethanol. *Process Biochemistry*, 1;51(10):1636-43, 2016.
 25. Sirohi P, Verma H, Singh SK, Singh VK, Pandey J, Khusharia S, Kumar D, Teotia P, Kumar A. Microalgal Carotenoids: Therapeutic Application and Latest Approaches to Enhance the Production. *Current Issues in Molecular Biology*, 9, 44(12):6257-79, 2022.
 26. Marino T, Casella P, Sangiorgio P, Verardi A, Ferraro A, Hristoforou E, Molino A, Musmarra D. Natural beta-carotene: A microalgae derivate for nutraceutical applications. *Chemical Engineering Transactions*, 79:103-8, 2020.
 27. Nisar, N.; Li, L.; Lu, S.; Khin, N.C.; Pogson, B.J. Carotenoid Metabolism in Plants. *Mol. Plant*, 8, 68–82, 2015.
 28. Bccresearch report, 2019
 29. Market Data Forecast report (2023-2028)