Dunaliella Salina Teod's Growth, Biopigment, and Lipid Content Are Affected By Physical and Dietary Stressors.

Dr. Puran Prabha Associate Professor Department of Botany, G. C. Narnaul, Haryana, India, dr.pprabha99@gmail.com

ABSTRACT

About half of the photosynthesis on earth is performed by the widespread group of photosynthetic organisms known as algae. Numerous different types of creatures fall under the umbrella name "alga." They can be either tiny, single-celled microalgae or bigger, more complicated macroalgae with many cells. They can be found all over the world in a variety of situations, including freshwater and marine habitats. Microalgae are autotrophic microorganisms that use light energy and inorganic nutrients to create pigments, lipids, proteins, carbohydrates, and other important biomass molecules. One of the most significant natural resources on the planet is algae. They support the food chain for more than 70% of the world's biomass, and they contribute to about 50% of the photosynthetic activity worldwide (Wiessner et al., 1995). Algae have a significant part in phytoplankton, which accounts for the majority of aquatic productivity. In addition to being essential environmentally and economically, marine algae have been utilized for ages as food and medicine. Diverse marine algae species now produce extracts like agar, carrageenans, and alginates in addition to serving as a food source. These extracts have diverse applications in the food, medicinal, cosmetic, and industrial sectors.

INTRODUCTION

They support the food chain for more than 70% of the world's biomass, and they contribute to about 50% of the photosynthetic activity worldwide (Wiessner et al., 1995). Algae have a significant part in phytoplankton, which accounts for the majority of aquatic productivity. In addition to being essential environmentally and economically, marine algae have been utilized for ages as food and medicine. Diverse marine algae species now produce extracts like agar, carrageenans, and alginates in addition to serving as a food source. These extracts have diverse applications in the food, medicinal, cosmetic, and industrial sectors. Halophilic biflagellated microalga include Dunaliella. A range of unicellular species that flourish in hypersaline, marine, and fresh water habitats make up the genus Dunaliella, which is categorized under the class Chlorophyceae, order Volvocales (Avron and Ben-Amotz, 1992). According to Garcia et al. (2007), Dunaliella is the only eukaryotic and photosynthetic organism that can thrive in a wide range of salt concentrations, from 0.05 M to saturation (5.0 M). The majority of the primary production in hyper-saline environments globally is carried out by this biflagellate unicellular alga (Oren, 2005). The first instance of Dunaliella was documented by Michael Felix Dunal in a salt evaporation pond in southern France in 1838.

Carotenoids perform several functions in microalgae. They are involved in light harvesting, but also contribute to stabilize the structure and aid in the function of photosynthetic complexes. The pigmentation properties of carotenoids have granted to some of them extensive application in the food and feed industry (Borowitzka and Borowitzka, 1988 and Todd Lorenz and Cysewski, 2000). In algae and higher plants, carotenoids play multiple and essential roles in photosynthesis. They contribute to light harvesting, maintain structure and function of photosynthetic complexes, quench chlorophyll triplet states, scavenge reactive oxygen species, and dissipate excess energy (Demming-Adams and Adams, 2002). Carotenoids also participate in different types of cell signaling (Cogdell, 1978). They are able to signal the production of absicisic acid, which regulates plant growth, seed dormancy, embryo maturation and germination, cell division and elongation, floral growth and stress responses(Finkelstein, 2013). Because of their antioxidant properties, carotenoids also have application in cosmetics (Del Campo et al., 2000). A large number of research studies have been confirmed the benefits of carotenoids to health and their use for this purpose is growing rapidly.

There has been a wide range of studies carried out on lipid induction techniques in microalgae such as the use of nutrients stress, including nitrogen and/or phosphorus starvation, light irradiation, pH, temperature, heavy metals and other chemicals. The microalgae also have applications in environmental biotechnology since they can be used for bioremediation of wastewater and to monitor environmental toxicants. Algal biomass produced during wastewater treatment may be further valorized in the biofuel manufacture. It is anticipated that the high microalgal lipid potential will force research towards finding effective ways to manipulate biochemical pathways involved in lipid biosynthesis and towards cost effective algal cultivation and harvesting systems, as well.

Microalgae have been explored for their bioactive compounds with promising applications. Considering the present status of widely used treatment therapies and their limitations to tackle their adverse effects, the application of bioactive compounds derived from algae will prove beneficial and much more effective as compared with traditional treatment methods. Basic research is micro algal field needed to find new strains amenable to fast growth at high cell densities and producing cells with a high content of desired products (Richmond, 2004). Algal strain selection and process optimization may be the key areas for future development (Wijffels, 2007). Thus the green alga Dunaliella salina isolated from Sambhar salt lake, Jaipur (Rajasthan) was selected to the present study.

The optimization of the biomass yield is the main factor in large scale cultivation of microalgae. Thus, it is necessary to understand the behavior of algal species under different environmental factors that determine the different growth parameters. The study of the interactions between these factors and growth modeling parameters allows finding the optimal conditions for selected species in large-scale productivity. Overall, the growth and biochemical composition of microalgal populations depends on many factors, the most important of which are nutrient availability, temperature and light (Carvalho and Malcata, 2003; Bardhan et al., 2008; Samuel et al., 2010 and Pandey et al., 2011).With the advancement in this field in recent years, the production of bio-organic compounds such as β-carotene is formed in many countries. It has been reported that large-scale production of β-carotene and Lipid induction is controlled by numerous stress factors like extreme light intensity, high salinity, temperature and availability of nutrients. D.salina contains up to 10% β –carotene on dry weight basis when grew under stress conditions including; high salinity (Loeblich, 1982), low growth temperature (Abdullaev and Semenenko, 1974), far red light and nutrient limitation conditions (Ben-Amotz, 1987;Borowitzka and Borowitzka1988b). An optimum light requirement varies with the microalgal species, and several parameters need to be considered in selecting the right combination when using artificial light in view of the overall energy balance considerations. It may be feasible to use high light intensities for enhanced production of lipids, biomass as well as suitable fatty acid profile for improving biofuel potential. Lipids productivityis therefore found to be influenced under high light stress.

Suitable nutrient medium for the culturing of algae is happened to be the prime requirement of biomass production. Inorganic constituents of the media are generally said to be responsible for the growth and morphology of the various algae in nature as well as in laboratory. It is well known fact, that the growing algae generally absorb many important minerals and trace elements from its habitat and naturally chelate them with the protein molecules found in it (Henrikson, 1989). Inorganic constituents of a medium i.e. nitrogen, potassium, sulphur, magnesium, zinc, cobalt, copper, boron and molybdenum were play an important role in growth and morphology of various algae in laboratory (Chu, 1942 and Allen, 1968). Many media have been invented for culturing different algae but a single medium has not been identified as the best one. However, different suggested media have inflicted differential impacts on the growth of the algae.

Microalgae such as *Dunaliella salina* require a number of macro and micronutrients for growth. Nutrient availability has a significant impact on growth and propagation of microalgae and broad effects on their lipid and Fatty acids composition. Environmental stress condition when nutrients are limited, invariably cause a steadily declining cell division rate. The supply of the limiting nutrients is eliminated under the state of nutrient starvation, where the growth of microalgae is affected, and the cells modify its physiological functions with accumulation ofcertain molecules to withstand in stress conditions.

CONCLUSION

According to Markou and Nerantzis (2013), microalgae are autotrophic organisms that create biomass rich in value-added products including lipids, carbohydrates, proteins, and pigments by using light energy and inorganic nutrients. In addition to being extensively studied for their lipid content, microalgae also produce metabolites that are widely used as food additives in the pharmaceutical industry, including carotenoids (lutein, zeaxanthin, and astaxanthin), long-chain polyunsaturated fatty acids (LC-PUFA), and vitamins (Pal et al., 2011). The increase in rich reserves of highly valuable metabolites is what attracts interest in the large-scale cultivation of microalgal biomass. In accordance with (Clarens et al., 2010; Norsker et al., 2011; Soratana and Landis, 2011). The group of algae is extremely diverse and heterogeneous, and its members inhabit a wide range of habitats. Every living thing has the innate ability to react to stimuli or changes in its environment. As a result, changes in external conditions can be classified as either stressors or limiting factors based on the reaction that the cell has to the observed change. For the sake of simplicity, we define a limiting factor as one that controls the rate of a biochemical reaction or rate of development and that a change in its level will cause a change in the rate without the need for an acclimation phase.

Such alterations are aimed to help balance efficiently the absorption of excitation energy and the production of reducing power (NADPH) and chemical energy (ATP) with their utilization for growth and cell maintenance. Inability to maintain this balance due to excess excitation of the photosynthetic reaction centers may result in the production of toxic oxygen species that may lead to photo- oxidative death. As implied, many of the stress responses and adaptive process are associated with the photosynthetic apparatus. Indeed, microalgae of different origins have a tendency, albeit with certain exceptions, to resemble each other in terms of cell composition, particularly in the relative amounts of crude protein, lipids, and carbohydrate that they contain when grown under more or less optimal growth conditions. For a single species, on the other hand, the variation in cell composition may differ many fold, according to the culture conditions under which it is grown. For example, *Chlorella sp*., *Botryococcus braunii*, and *Dunaliella salina*, which are all classified under Chlorophyceae, Volvocales, show typical biochemical composition: 30–50% proteins, 20–40% carbohydrate and 8–15% of lipids under favorable environmental conditions. These species, however, can accumulate under unfavorable environmental conditions up to 80% of fatty acids, 80% of hydrocarbons, and 40% of glycerol, respectively, on the basis of the dry weight.

Clearly, environmental factors, particularly light, temperature, nutrient status, and salinity, not only affect photosynthesis and productivity of cell biomass, but also influence the pattern, pathway and activity of cellular metabolism and thus dynamic cell composition. The effects on the latter have extensive biotechnological implications and consequences. Biomass production in turn was depended upon several factors. The growth of any unicellular alga is a manifest of an increase in size followed by the division of the cells, which is said to be influenced by the nutritive medium, light quality and photoperiods accompanied by temperature.

It is very interesting that growth of algae is effected by chemical composition of various inorganic media used in the culture. There are many elements like sodium, nitrate, chloride, phosphate, sulphate, magnesium, and carbonate that have significant role in growth of the algae (Arnon and Wessel, 1953;O'Kelley, 1968) and lack of any of this element may reduce the growth of algal cultures. Aslan and Kapdan (2006) showed that phosphate removal for *C. vulgaris* in artificial wastewater decreased when light limitation became an issue. Estimation of microalgae growth is generally expressed in: increase in optical density, protein, pigment over a period of time (Becker, 1994, Junior *et al.*, 2007).

According to Donald, (1963) algal cultures could be maintained for periodsof several weeks by supplementing the nutrient containing minimal amount of certain salts. In the initial stage more and more available nutrients are utilized for theformation of cell structure and reserved food material, as the cells are increasing in number. Rapidly growing microalgal cultures exhibit higher protein content (Sayeghand Montagnes, 2011).

In the present investigation**,** nutrient requirements of *Dunaliella salina* has been worked out employing four inorganic media varying in their chemical composition and pH. Among various inorganic media tested for *D.salina*, the best growth was found in Artificial Sea Water medium followed by De Walne's, Modified Johnson's respectively and least growth was observed in Bold's Basal medium. Highest growth rate (0.326 divisions/day) was calculated in Artificial Sea Water medium. While least growth rate (0.186 divisions/day) was recorded in the Bold's Basal medium. Here the optical density and dry weight of the alga also supported these findings.

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