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Industrial Production of Microalgae *Arthrospira (Spirulina) platensis* in the Central Iran

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ABSTRACT

Spirulina platensis (Arthrospira platensis) is the most common microalgae belong to blue-green algae. It grows in water, can be harvested and processed easily. In many countries, it is used as human food as an important source of protein, vitamin and supplement to aquaculture diets. This group of microalgae represents one of the most promising sources for new products and applications. This work addresses the assessing culture of Spirulina platensis under open ponds and laboratory-scale (polyethylene bags) in the Danesh Pazhohan Sabz Bonyan Qeshm Company (Iran). In conclusion, we observed that density ranged from 4521 to 7500 (cellml^{-1}) in lab condition and 5302 to 9981 (cellml^{-1}) in open ponds. The protein levels measured at the end of the experiment were 37 and 55% for Lab and ponds cultures, respectively. As a result, in this study, we obtained the most biomass production in the open ponds.

Key words: *Microalgae, Spirulina, Open pond, Commercial Production, Cultivation Methods.*

INTRODUCTION

Spirulina platensis is a multicellular, filamentous cyanobacterium, that is commercially and biotechnologically important due to its high content of protein (up to 70%), pigments (e.g. phycocyanin, allophycocyanin, chlorophyll a, carotenoid and phycoerythrin), essential fatty acids (γ-linolenic acid) [1,2] vitamin B_{12} and minerals[3]. Also, *Spirulina* has some medicinal uses such as enhanced brain function, behavior and learning, protect from radiation, powerful anticancer properties and detoxification support[4,5]. *Spirulina* can colonize environments that are unsuitable for many other organisms, forming populations in freshwater and brackish lakes and some marine environments, mainly alkaline saline lakes[6,7].

The large-scale production of *Spirulina* biomass depends on many factors including of nutrient availability, temperature and light. These factors can influence the growth of *Spirulina* and the composition of the produced biomass by causing changes in metabolism, which considerably modify the time course of the accumulation of the main biomass components[8].

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Carbon is the principal nutrient required by *Spirulina*, and in alkaline lakes this organism is the dominant species because of the presence of high concentrations of sodium carbonate. Vonshak[9] has shown that, after labor, the second major cost in *Spirulina* biomass production is the cost of nutrients, principally the carbon source. Amount of production annually *Spirulina* is different in countries include Japan, China, Taiwan, Thailand, Hawaii, Coba (1100, 500, 480, 170, 500 and 40 tone, respectively[10]. The optimal temperature for *Spirulina* is in the range of 35-38ºC. In addition, *Spirulina* requires relatively high pH values between 9-9.8 [11], which effectively inhibit the contamination of most algae in the culture. In this present research an attempt has been made to production of *Spirulina* in open pond and polyethylene bags. During production cell growth is followed by measuring the optical density of the culture medium. The industrial scale production of biomass the energy consumption require for mass transfer and the arrangement of light receiving surface of algal suspension must be require to its minimum possible level[12].

MATERIALS AND METHODS

The cyanobacterium used in this study was *Spirulina platensis* strain PCC 7345 (Pastur Company Cyanophyta). We in laboratory period tested two BG_{11} and Zarrouk medium on *Spirulina platensis* but, finally Zarrouk's medium[13,14], being used to prepare and maintain the cultures. *Spirulina platensis* was cultivated under two different culture conditions open ponds (Dimension: 85×185 cm, 50 cm depth) and in laboratory in the Danesh Pazhohan Sabz Bonyan Qeshm Company (Iran). Growth of *Spirulina platensis* was measured daily by taking optical density of culture by using Spectrophotometer 166, as well as counting density with Sedgwick-Rafter slide. Identification was done on the basis of the morphological characters of the algae with a Zeiss Axioster Plus research microscope. Protein was estimated by the methods as described by Lowry *et al*[15]. The cultivation conditions were agitation, 160 rpm, temperature, 30ºC and pH of 9-10. Chemicals were added according to Zarrouk formula. The chemicals were added just after every harvesting by calculation. *Spirulina* cells were harvested firstly with 55 μ-mesh nylon cloth. The filter was dried at 105ºC for 24 h, and weighed. Harvesting was performed by pumping the cultures into. The filtrate was pumped back into the ponds, while the algal slurry was pumped to a Niro minor spray drier (Niro, Denmark) without further washing.

RESULTS AND DISCUSSION

Spirulina platensis was cultured in lab condition and open ponds. The color of media gradually changed to the dark green in 15 days which developed further due to the lush growth of *Spirulina.* Morphologically trichomes were composed of cylindrical cells with 2-10 μm diameters and 1 mm in length (**Figure 1**).

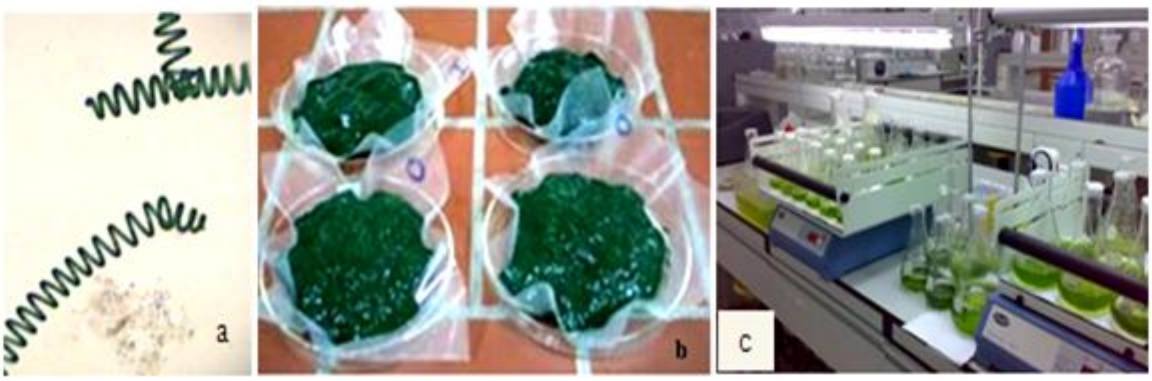

Fig. 1: a) Structure of *Spirulina platensis* b) Collected biomass of *Spirulina platensis* in the open ponds c) Culture in laboratory

The optical density of each measured by regular intervals of 2 days. The OD of culture was 1.3 to 22.6 from 1st to 20th day, as well as, density based on counting by Sedgwick-Rafter ranged from 4521 to 7500 cellml^{-1} in lab condition and 5302 to 9981 cellml^{-1} in open ponds (**Figure 2**), which minimum growth rate represented in laboratory condition. In general, the highest density observed in eleven days in open ponds. The OD of cultures which were kept under laboratory conditions was ranged 1.23 to 16.7 from 1st to 20th day.

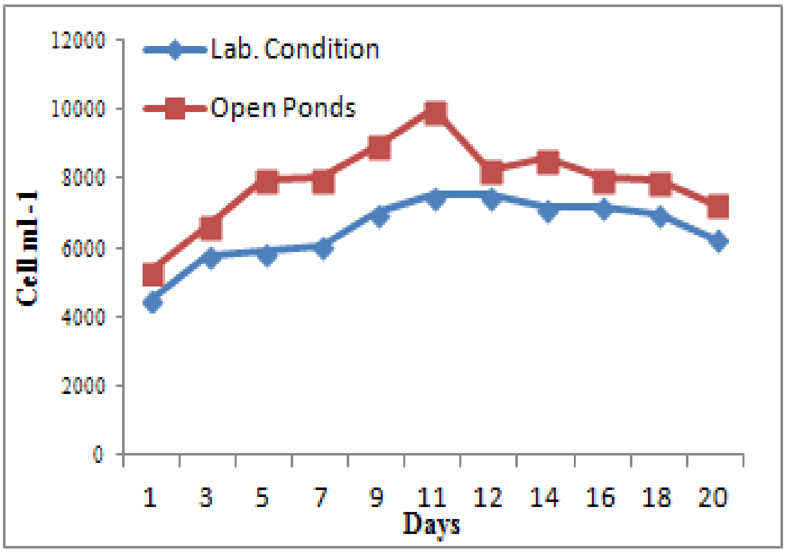

Fig. 2: Density of *Spirulina* in the both medium for 20 days

The protein in lab condition was 37%, while in open pond was 55% (**Figure 3**).

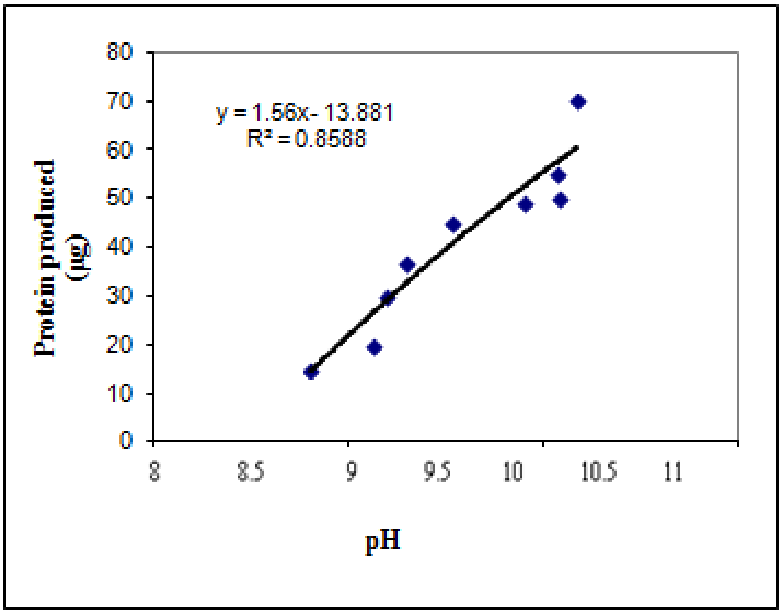

Fig. 3: Correlation between pH and protein produced (µg) in open ponds condition

Also, amount of produced biomass in the both culture medium are present in Figure 4, which in this study, we obtained most biomass in fifteen day (4 gl^{-1}) in the open ponds, as well as, biomass lowest was obtained in third day at lab condition (0.5 gl^{-1}).

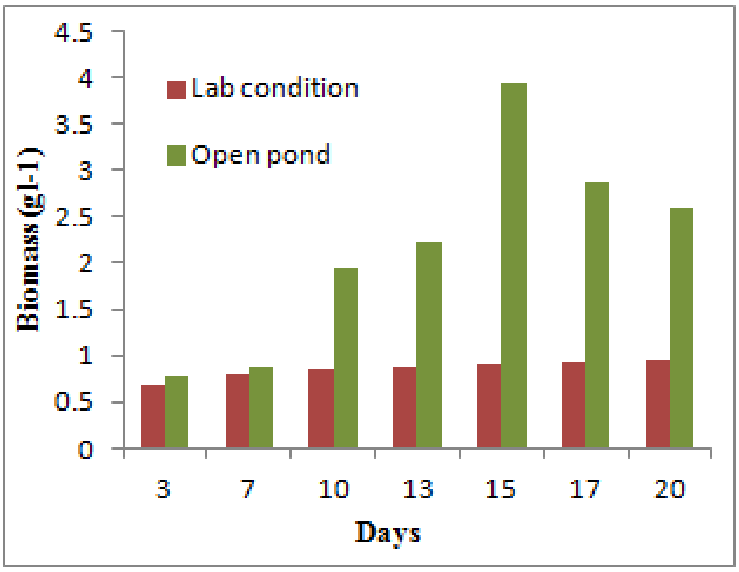

Fig. 4: Comparing of produced biomass in the lab condition and open ponds

Spirulina platensis is a multicellular, filamentous cyanobacterium that is commercially and biotechnologically important due to valuable properties. *Spirulina platensis* was cultured under two different conditions to explore optimum condition, which the open ponds were better condition for industrial culture. The growth parameters like temperature (35-38ºC) and pH (9.5) controlled in both condition. The best pH for *Spirulina* growth was ranged between 9 and 10 especially in BG11 medium (**Figure 5**). Among conditions, the optimum growth performance of *Spirulina platensis* was observed in open ponds. Richmond *et al*.[16], observed that the low temperature not suitable for the growth of *Spirulina*[16].

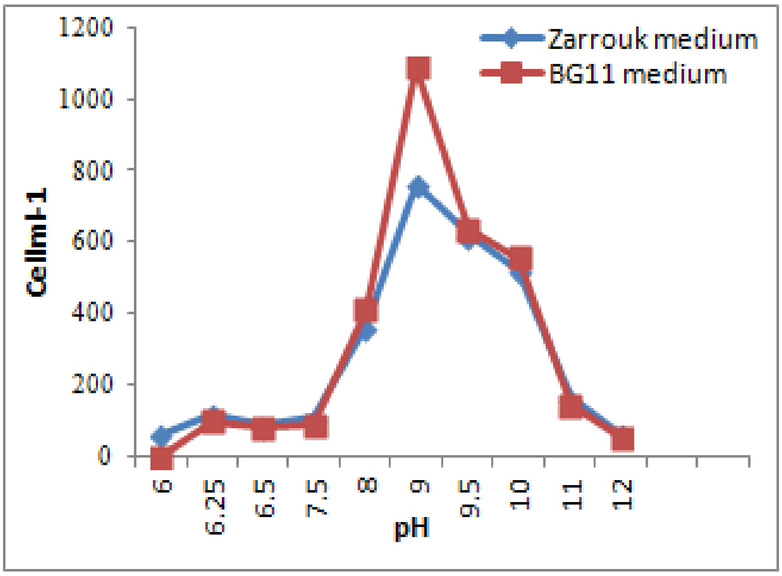

Fig. 5: Comparing of cell density under different pH in the both culture medium.

The first harvesting was done after 20 days of culture and further harvesting was performed after every thirtieth days. It is very important to explain that the helical shape of *Spirulina platensis* is changed to spiral shape in different media (BG_{11} and Zarrouk). Also, in this study we observed that Zarrouk medium was better than BG11 for cell growth, in *Spirulina* (**Figure 6**).

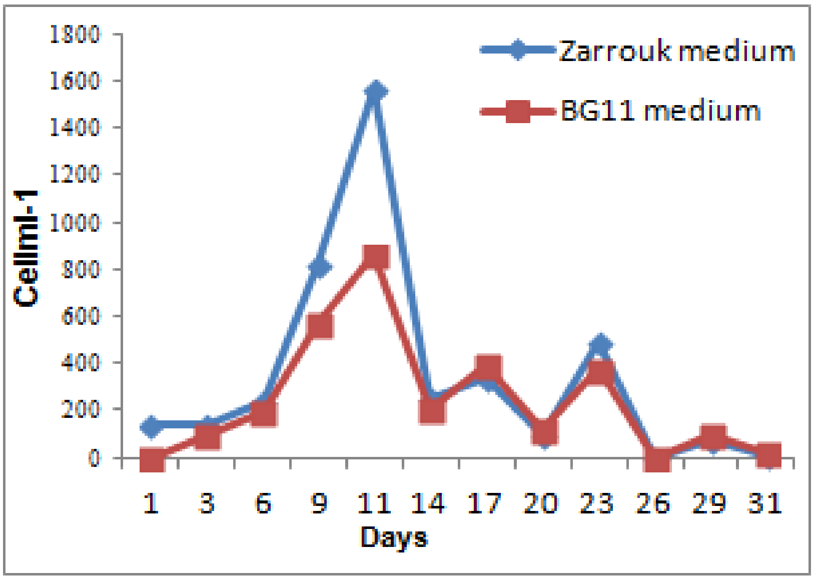

Fig. 6: Comparing of cell density under different culture medium.

These changes are due to hydration or dehydration of oligopeptides in the peptidoglycan layer. Reasonably, high correlation coefficients were obtained between protein produced and pH. Our results show that pH determination can be used as an indicator of algae growth, which these results was similar to Pelizer *et al*[17]. In addition, amounts of protein in open ponds was more than of Lab condition, which this result was resemble to finding of Soni *et al*[18].

CONCLUSION

However, most producers have been confronted with several problems before they achieved economic success in *Spirulina*. These problems relate mainly to open ponds production and harvest efficiency. Another major problem is the maintenance of stable and high quality in open ponds grown *Spirulina*. Results showed that, industrial culture in open ponds was good for successful cultivation of *Spirulina platensis* under semi-arid climatic condition of Qeshm Island. In conclusion, cooperation between researchers in the academia and those with rich experience in actual open ponds mass culture can bring signification improvement in this area and bring down the cost of production so that many more people can benefit from the health food qualities of *Spirulina.*

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