

Effect of different culture media on growth of *Spirulina platensis*

Preeti Soni¹, Gajendra Pal Singh², Vishal Sharma³

¹ Scholar, Department of Botany, University of Rajasthan, Jaipur, Rajasthan, India

² Professor, Department of Botany, University of Rajasthan, Jaipur, Rajasthan, India

³ Department of Sciences, Vivekananda Global University, Jaipur, Rajasthan, India

Abstract

Spirulina platensis is a filamentous cyanobacterium recognized for its high nutritional value and extensive applications in the food, pharmaceutical, cosmetic, and aquaculture industries. Due to its rich composition of proteins, vitamins, essential amino acids, pigments, and antioxidants, large-scale cultivation of *Spirulina* has gained considerable commercial importance. The present study was conducted to evaluate the influence of different culture media on the growth performance of *Spirulina platensis* under controlled laboratory conditions. The organism was cultivated in various nutrient media, and growth was monitored by measuring optical density at regular intervals. Significant variations in growth rate were observed among the tested media, indicating that nutrient composition strongly affects the metabolic activity and nutrient assimilation efficiency of the cyanobacterium. Among the media tested, certain formulations supported superior growth due to the availability of balanced macro- and micronutrients essential for photosynthesis and cellular development. The findings of this study demonstrate that optimization of culture media is a key factor for enhancing growth of *Spirulina platensis*. Furthermore, the study highlights the potential for developing cost-effective cultivation strategies for sustainable commercial production and biotechnological applications.

Keywords: *Spirulina platensis*, culture media, biomass production, optical density, cyanobacteria cultivation

Introduction

Algae are increasingly recognized as sustainable bioresources due to their rapid growth, high photosynthetic efficiency, and ability to synthesize valuable biomolecules such as proteins, lipids, carbohydrates, and pigments under diverse environmental conditions (Sánchez-Bayo *et al.*, 2020; Ekpan *et al.*, 2025) [18, 39]. Among these, cyanobacteria, particularly *Spirulina* (*Arthrospira platensis*) are of significant importance due to their high protein content, rich phycobiliprotein pigments, and exceptional nutritional value (Vonshak, 1997; Falquet & Humi, 1997; Becker, 2013) [9, 20, 43].

The growth, metabolism, and biochemical composition of *Spirulina* are strongly influenced by environmental and nutritional factors, among which culture medium composition plays a central role (Richmond, 2004; Vonshak & Richmond, 1988) [35, 45]. Culture media supply essential macro- and micronutrients, inorganic carbon sources, and buffering systems required for photosynthesis and biomass production. Variations in nutrient availability—particularly nitrogen, phosphorus, bicarbonate, and trace elements—significantly affect growth rate, pigment synthesis, and overall productivity (Chu, 1942; Colla *et al.*, 2007; Nyabuto *et al.*, 2015; El-Sheekh *et al.*, 2021) [13, 15, 19, 31].

Microalgae and cyanobacteria exhibit dynamic physiological responses to nutrient fluctuations. Nutrient-rich media promote protein synthesis and pigment accumulation, whereas nutrient limitation often induces stress responses leading to increased lipid and carbohydrate accumulation (Cordeiro *et al.*, 2017; Sánchez-Bayo *et al.*, 2020) [16, 39]. These responses highlight the importance of media optimization in regulating metabolic pathways and enhancing desired biochemical outputs.

Across both classical and modern studies, culture medium composition has been identified as the primary driver of biomass productivity and biochemical characteristics in *Spirulina platensis*. Foundational formulations such as Zarrouk's medium demonstrate that variations in nitrogen, phosphorus, salinity, and trace metals significantly influence physiological performance (Zarrouk, 1966; Vonshak & Richmond, 1988; Ajayan, 2023) [4, 45, 47]. Comparative studies further show that while Zarrouk's medium generally supports maximum biomass and pigment production, alternative media provide moderate growth, and low-cost or wastewater-based media offer economically viable but variable productivity (Ilavarasi *et al.*, 2011; Salunke *et al.*, 2016; Raoof *et al.*, 2006; Madkour *et al.*, 2012) [24, 27, 34, 38].

Role of Key Nutrients in Media Composition

Nitrogen is the most critical macronutrient influencing *Spirulina* growth and metabolism. High nitrogen availability enhances biomass accumulation, protein synthesis, and phycocyanin production, whereas nitrogen limitation reduces growth and induces lipid accumulation as a stress response (El-Sheekh *et al.*, 2021; Cordeiro *et al.*, 2017) [16, 19]. Continuous culture studies confirm that nitrogen levels directly regulate macromolecular composition and nutrient use efficiency (Khannapho *et al.*, 2021) [25] as shown in Figure 1.

Phosphorus and Mineral Balance: Phosphorus plays a vital role in ATP synthesis, energy metabolism, and cell division. Along with essential ions such as sodium, potassium, calcium, and magnesium, it contributes to structural stability and metabolic activity. Deficiency in phosphorus leads to reduced growth and decreased chlorophyll and phycobiliprotein content (Cogne *et al.*, 2003; Abobaker *et al.*, 2016) [2, 14].

Trace Elements and Biochemical Modulation: Trace elements such as iron (Fe), zinc (Zn), and copper (Cu) are crucial for enzymatic activity and pigment biosynthesis. Iron enhances chlorophyll and phycocyanin synthesis, zinc supports metabolic enzymes, and copper, at optimal levels,

improves metabolic balance but becomes toxic at higher concentrations. Optimized trace element supplementation has been shown to increase phycobiliproteins, antioxidant activity, and biomass yield (Frontasyeva, 2009; Akbarnezhad *et al.*, 2020) [5, 22].

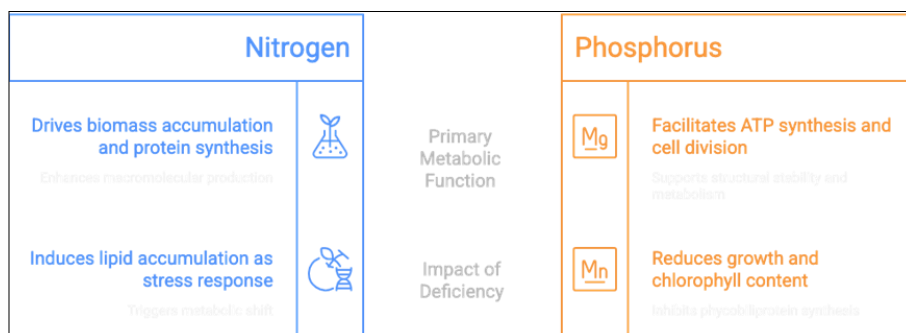


Fig 1: Role of Nutrients in Media Composition

Influence of Media on Biochemical Composition

Culture media directly regulate the biochemical profile of *Spirulina*:

- **Proteins:** Increased under high nitrogen conditions, reaching up to 60–70% of dry weight; reduced under nutrient stress.
- **Lipids:** Accumulate under nitrogen or phosphorus limitation, important for biofuel applications (Cordeiro *et al.*, 2017; Ekpan *et al.*, 2025) [16, 18]
- **Carbohydrates:** Serve as storage compounds and increase under stress conditions
- **Phycobiliproteins:** Highest under balanced nutrients and moderate light; reduced under nutrient deficiency (Khatoun *et al.*, 2018) [26]

Interaction with Environmental Factors

Although media composition is central, its effects are strongly influenced by environmental conditions. Light intensity interacts synergistically with nutrient availability—high light enhances photosynthesis but requires sufficient nutrients, while light limitation restricts growth even in nutrient-rich media (Vonshak *et al.*, 2014; Benyamin, 1993) [10, 44]. Similarly Optimal growth occurs at alkaline pH (~9–10) typical of *Spirulina* habitats and inoculum size also affects lag phase and adaptation with higher inoculum influences faster exponential phase while low inoculum extended lag phase (Ale *et al.*, 2014) [6]

Environmental stress factors such as UV radiation, electric fields, and oxidative stress further modify biochemical responses. Controlled stress can enhance valuable metabolites like pigments, whereas excessive stress reduces overall productivity (Vonshak&Torzillo, 2003) [46].

Industrial and Biotechnological Significance

Optimization of culture media is essential for large-scale *Spirulina* cultivation. Proper nutrient formulation enhances biomass yield, protein content, and pigment production, supporting applications in functional foods, pharmaceuticals, animal feed, and biofuel production (Borowitzka&Vonshak, 2017; Abdel-Wareth *et al.*, 2024 [1, 12]; AlFadhly *et al.*, 2022). Additionally, development of cost-effective media formulations enables sustainable

industrial-scale production without compromising productivity.

In this context, the present study aims to evaluate the effect of different culture media (Zarrouk's, BG-11, BBM, and CHU-10) on the growth performance of *Spirulina platensis* using optical density measurements and biochemical analysis. Understanding these differences is essential for optimizing culture conditions for both laboratory and industrial applications.

Culture medium composition is the primary controlling factor in *Spirulina* biotechnology. Balanced macro- and micronutrients promote high biomass and protein-rich cultures, whereas nutrient limitation induces metabolic shifts toward lipid and carbohydrate accumulation. The interaction between media composition, light, and environmental stress ultimately determines growth performance and biochemical output. Therefore, precise optimization of culture media is crucial for maximizing productivity and tailoring metabolite profiles for specific industrial applications.

Materials and Methods

Isolation and Maintenance of *Spirulina platensis*

Water samples containing mixed algal populations were collected from Jal Mahal Lake, Jaipur (Rajasthan), India and transported immediately to the laboratory. The samples were centrifuged at 2000–3000 rpm for 5–10 minutes to concentrate algal biomass. Isolation of *Spirulina platensis* was carried out using enrichment culture, serial dilution, agar plating, and micropipette isolation methods following standard protocols (Andersen, 2005; Singh & Dhar, 2020) [8, 40]. The isolated organism was identified morphologically by observing unbranched helical trichomes under a light microscope (Vonshak, 1997) [43]. Pure cultures were maintained in sterile Zarrouk's medium under controlled laboratory conditions at 27 ± 3°C with a 16:8 h light–dark cycle and light intensity of 2000–3000 lux. Subculturing was carried out every 10–15 days under aseptic conditions.

Experimental Setup

To evaluate the effect of different nutrient compositions on growth and pigment production, *Spirulina platensis* was cultured in four different media: Zarrouk's Medium, BG-11 Medium, Bold's Basal Medium (BBM), Chu-10 Medium. The media were prepared according to standard

compositions and sterilized at 121°C for 15 minutes. Equal volumes of actively growing inoculum were transferred into sterile 250 mL conical flasks containing the respective media and incubated medium under controlled laboratory conditions at $27 \pm 3^\circ\text{C}$ with a 16:8 h light–dark cycle and light intensity of 2000–3000 lux. The experiment was conducted in triplicate under identical culture conditions. Observations were recorded weekly to assess changes in growth.

Growth Analysis

Growth was estimated by measuring optical density at 680 nm (OD_{680}) using a UV–Visible spectrophotometer. Increased OD values indicated enhanced biomass accumulation.

Results

The growth of *Spirulina platensis* was evaluated over a period of five weeks using optical density (OD) measurements in four different culture media: Zarrouk's medium, BG-11, BBM, and CHU-10.

A gradual increase in OD (optical density) was observed in all media, indicating active growth; however, the extent of growth varied significantly depending on the nutrient composition of the medium.

During Week I, Zarrouk's medium exhibited the highest mean OD (0.212 ± 0.0116), followed by BG-11, BBM, and CHU-10. Statistically significant reductions in growth were observed in BBM and CHU-10 ($p < 0.01$), while BG-11 showed a moderate decrease ($p < 0.05$).

From Week II to Week III, a sharp increase in OD indicated the exponential growth phase (0.803 ± 0.0206 and 1.583 ± 0.0290 , respectively). In contrast, CHU-10 exhibited the lowest growth, with a reduction reaching 40.48% by Week III.

In Week IV and Week V, growth continued to increase, reaching maximum OD values in Zarrouk's medium (2.129 ± 0.0369 and 2.582 ± 0.0401 , respectively). The reduction in growth compared to Zarrouk's medium was highest in CHU-10 (-54.98%), followed by BBM (-40.87%) and BG-11 (-27.83%), all statistically highly significant ($p < 0.001$) as shown in figure 2.

Overall, the order of growth performance was:

Zarrouk's > BG-11 > BBM > CHU-10

Discussion

The present study clearly demonstrates that culture medium composition is a critical factor influencing the growth and biomass productivity of *Spirulina platensis*, as reflected by the progressive increase in optical density (OD) across all treatments with significant variations among media.

Zarrouk's medium consistently supported the highest growth throughout the experimental period. This superior performance is primarily attributed to its high bicarbonate concentration, alkaline pH, adequate nitrogen supply, and rich mineral composition, which together provide optimal conditions for photosynthesis and biomass accumulation. *Spirulina* thrives in bicarbonate-rich environments where inorganic carbon enhances carbon fixation (Zarrouk, 1966; Richmond *et al.*, 1982; Vonshak, 1997) [36, 43, 47]. Similar observations were reported by Richmond (2004) and Vonshak (1997) [35, 43], emphasizing the importance of carbon availability, while Aiba and Ogawa (1977) [3] further

demonstrated that *Spirulina* achieves higher growth yield under nutrient-rich, non-limiting conditions.

BG-11 medium supported moderate growth but remained significantly lower than Zarrouk's medium. This can be explained by its lower bicarbonate content, limited carbon availability, and lack of high alkalinity, as it is primarily designed for freshwater cyanobacteria rather than *Spirulina* (Rippka *et al.*, 1979) [37]. The observed reduction (~27% by Week V) supports this limitation.

BBM showed further reduced growth, likely due to its formulation for green algae rather than cyanobacteria, resulting in lower alkalinity and suboptimal nutrient balance. Bischoff and Bold (1963) [11] originally developed BBM for chlorophytes, and studies by Markou and Nerantzis (2013) [29] confirm that non-optimized media significantly reduce *Spirulina* biomass due to stress conditions.

CHU-10 exhibited the lowest growth, with a drastic reduction (~55% by Week V). This poor performance can be attributed to insufficient bicarbonate, lower nutrient concentration, and unsuitable pH conditions. Chu (1942) [13] designed this medium for freshwater algae, and it does not meet *Spirulina*'s physiological requirements. Similar findings by Richmond (2004) and Markou *et al.* (2012) [30, 35] indicate that *Spirulina* cannot grow efficiently without adequate carbon and alkalinity.

The growth pattern observed—lag phase (Week I), exponential phase (Weeks II–III), and late exponential phase (Weeks IV–V)—follows the typical microbial growth curve and is consistent with previous reports (Pelizer *et al.*, 2003; Vonshak & Richmond, 1988; Danesi *et al.*, 2002) [17, 32, 45].

Nutrient availability plays a crucial role in determining growth performance. Nitrogen influences protein synthesis and biomass accumulation (Colla *et al.*, 2007) [15], while bicarbonate enhances carbon fixation and maintains alkaline pH (Richmond *et al.*, 1982; Magwell *et al.*, 2023) [28, 36]. Additionally, environmental factors such as light intensity and temperature further influence growth dynamics (Danesi *et al.*, 2002; Torzillo *et al.*, 1991) [17, 42]. Medium optimization has been shown to significantly enhance productivity (Raouf *et al.*, 2006; Madkour *et al.*, 2012) [27, 34], and recent studies confirm that modifying medium composition can improve both biomass and pigment production (Freire Balseca *et al.*, 2024) [21].

The present findings are in strong agreement with earlier studies reporting maximum growth in Zarrouk's medium and reduced growth in alternative media (Soni *et al.*, 2019; Rajasekaran *et al.*, 2016; Gami *et al.*, 2011; Salunke *et al.*, 2016) [23, 33, 38, 41].

From an industrial perspective, although Zarrouk's medium ensures maximum productivity, its high cost remains a limitation. Therefore, there is increasing emphasis on developing cost-effective alternative media without compromising nutrient balance and growth efficiency (Raouf *et al.*, 2006; Madkour *et al.*, 2012) [27, 34].

In conclusion, the study highlights that culture medium composition is a key determinant of *Spirulina* growth, with Zarrouk's medium providing the most favorable conditions, while CHU-10 is the least suitable. These findings emphasize the importance of medium optimization for efficient laboratory and large-scale cultivation

Summary

All media supported *Spirulina* growth, but efficiency varied significantly. Zarrouk's medium produced the highest biomass yield. Growth decreased in the order: Zarrouk's > BG-11 > BBM > CHU-10. Statistical analysis confirmed significant differences across media. Carbon availability and nutrient composition were the key controlling factors.

Conclusion

This study confirms that Zarrouk's medium is the most effective medium for the cultivation of *Spirulina platensis* due to its optimal nutrient balance, high bicarbonate concentration, and suitable alkaline conditions. BG-11 can be used as an alternative for moderate growth, while BBM and CHU-10 are comparatively unsuitable for *Spirulina* cultivation.

The results highlight that medium optimization is essential for maximizing biomass production, which is crucial for

commercial applications such as nutraceuticals, pharmaceuticals, and bioactive compound production. Future studies should focus on developing low-cost modified media that can match the efficiency of Zarrouk's medium while reducing production costs.

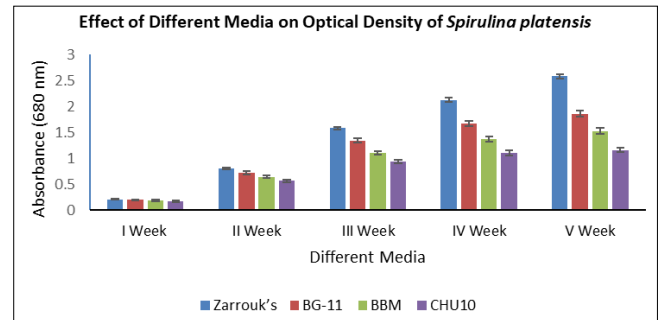


Fig 2: Different Media effect on Optical Density of *Spirulina platensis*

Table 1: Effect of different media on optical density (OD) of *Spirulina platensis*

Week	Zarrouk's (Control)	BG-11	BBM	CHU10
I	0.212 ± 0.0116	0.200 ± 0.0131 (-5.67%) *	0.184 ± 0.0162 (-13.23%) **	0.173 ± 0.0171 (-18.27%) **
II	0.803 ± 0.0206	0.725 ± 0.0287 (-9.63%) **	0.645 ± 0.0267 (-19.68%) ***	0.570 ± 0.0247 (-29.03%) ***
III	1.583 ± 0.0290	1.345 ± 0.0385 (-15.05%) ***	1.113 ± 0.0349 (-29.68%) ***	0.942 ± 0.0310 (-40.48%) ***
IV	2.129 ± 0.0369	1.676 ± 0.0453 (-21.29%) ***	1.379 ± 0.0516 (-35.25%) ***	1.107 ± 0.0424 (-48.01%) ***
V	2.582 ± 0.0401	1.863 ± 0.0539 (-27.83%) ***	1.527 ± 0.0580 (-40.87%) ***	1.163 ± 0.0445 (-54.98%) ***

Values expressed as Mean ± SD (n = 3) % change calculated relative to Zarrouk's medium, ns = non-significant; *p < 0.05; **p < 0.01; ***p < 0.001.

References

- Abdel-Wareth AA, Williams AN, Salahuddin M, Gadekar S, Lohakare J. Algae as an alternative source of protein in poultry diets for sustainable production and disease resistance: Present status and future considerations. *Frontiers in Veterinary Science*,2024;11:1382163.
- Abobaker HM, Elsalhin HE, Hasan HMA. Nitrogen, phosphorus and minerals (sodium, potassium and calcium). *International Journal of Current Microbiology and Applied Sciences*,2016;5(11):836–841.
- Aiba S, Ogawa T. Assessment of growth yield of a blue-green alga *Spirulina platensis* in axenic and continuous culture. *Journal of General Microbiology*,1977;102(1):179–182. <https://doi.org/10.1099/00221287-102-1-179>
- Ajayan KV. Microalgal cultivation: Cultivation media and its supplementary compounds. *Journal of Food and Agriculture Research*,2023;3(2):145–167.
- Akbarnezhad M, Shamsaie Mehrgan M, Kamali A, Javaheri Baboli M. Effects of microelements (Fe, Cu, Zn) on growth and pigment contents of *Arthrospira (Spirulina) platensis*. *Iranian Journal of Fisheries Sciences*,2020;19(2):653–668.
- Ale MT, Pinelo M, Meyer AS. Assessing effects and interactions among key variables affecting microalgae growth. *Preparative Biochemistry & Biotechnology*,2014;44(3):242–256.
- AlFadhly NK, Alhelfi N, Altemimi AB, Verma DK, Cacciola F. Trends affecting the growth and cultivation of genus *Spirulina*: An investigative review. *Plants*,2022;11(22):3063.
- Andersen RA. *Algal culturing techniques*. Elsevier Academic Press, 2005.
- Becker EW. *Microalgae: Biotechnology and microbiology*. Cambridge University Press, 2013.
- Benyamin AE. Light energy effect on *Spirulina platensis* growth rate (Doctoral dissertation), 1993.
- Bischoff HW, Bold HC. *Physiological studies IV. Some soil algae from enchanted rock and related algal species*. University of Texas Publication No. 6318, 1963.
- Borowitzka MA, Vonshak A. Scaling up microalgal cultures to commercial scale. *European Journal of Phycology*,2017;52(4):407–418.
- Chu SP. The influence of mineral composition of medium on growth of planktonic algae. *Journal of Ecology*,1942;30:284–325.
- Cogne G, Lehmann B, Dussap CG, Gros JB. Uptake of macrominerals and trace elements by *Spirulina platensis*. *Biotechnology and Bioengineering*,2003;81(5):588–593.
- Colla LM, Reinehr CO, Reichert C, Costa JAV. Nutraceutical production in *Spirulina*. *Bioresource Technology*,2007;98(7):1489–1493.
- Cordeiro RS, Vaz IC, Magalhães SM, Barbosa FA. Nutritional conditions and lipid production in cyanobacteria. *Anais da Academia Brasileira de Ciências*,2017;89(3 Suppl):2021–2031.
- Danesi EDG, Rangel-Yagui CDO, De Carvalho JCM, Sato S. An investigation of effect of replacing nitrate by urea in the growth and production of chlorophyll by *Spirulina platensis*. *Biomass and Bioenergy*,2002;23(4):261-269.
- Ekpan FM, Ori MO, Samuel HS. Effects of environmental factors and nutrient availability on the biochemical composition of algae for biofuel production. *Discover Applied Sciences*,2025;7(10):1209.

19. El-Sheekh M, Morsi H, Hassan L. Media optimization for *Spirulina platensis*. Egyptian Journal of Botany,2021;61(1):61–69.
20. Falquet J, Hurni JP. Nutritional aspects of *Spirulina*. Antenna Foundation, 1997.
21. Freire Balseca DA, Castro Reyes KS, Maldonado Rodríguez ME. Optimization of an alternative culture medium for phycocyanin production from *Arthrospira platensis* under laboratory conditions. Microorganisms,2024;12(2):363.
22. Frontasyeva M. Accumulation of trace elements by biological matrice of *Spirulina platensis*. ECOLOGICAL CHEMISTRY AND ENGINEERING S,2009;16(3):277-285.
23. Gami AS, Noheria A, Lachman N, Edwards WD, Friedman PA, Talreja D, et al. Anatomical correlates relevant to ablation above the semilunar valves for the cardiac electrophysiologist: a study of 603 hearts. Journal of Interventional Cardiac Electrophysiology,2011;30(1):5-15.
24. Ilavarasi A, Mubarakali D, Praveenkumar R, Baldev E, Thajuddin N. Optimization of various growth media to freshwater microalgae for biomass production. Biotechnology,2011;10(6):540-545.
25. Khannapho C, Phodee A, Paithoonrangsarid K, Hongsthong A, Meechai A, Cheevadhanarak S, et al. Effect of dilution rate in continuous cultures of *Arthrospira (Spirulina) platensis* C1 on nutrient use efficiency and macromolecular-and elemental compositions. Journal of Applied Phycology,2021;33(2):743-754.
26. Khatoon H, Leong LK, Rahman NA, Mian S, Begum H, Banerjee S, et al. Effects of light intensity and media on growth and pigment production of cyanobacteria. Bioresource Technology,2018;249:652–658.
27. Madkour FF, Nasr HS. Production and nutritive value of *Spirulina platensis* in reduced cost media. Egyptian Journal of Aquatic Research,2012;38(1):51–57.
28. Magwell PFR, Djoudjeu KT, Minyakaka E, Tavea MF, Fotsop OW, Tagnikeu RF, et al. Sodium bicarbonate (NaHCO₃) increases growth, protein and photosynthetic pigments production and alters carbohydrate production of *Spirulina platensis*. Current Microbiology,2023;80(2):63.
29. Markou G, Nerantzis E. Microalgae for high-value compounds and biofuels production: a review with focus on cultivation under stress conditions. Biotechnology advances,2013;31(8):1532-1542.
30. Markou G, Angelidaki I, Georgakakis D. Microalgal carbohydrates: an overview of the factors influencing carbohydrates production, and of main bioconversion technologies for production of biofuels. Applied microbiology and biotechnology,2012;96(3):631-645.
31. Nyabuto DK, Cao K, Mariga AM, Kibue GW, He M, Wang C. Environmental factors affecting growth and biochemical composition of *Spirulina*. African Journal of Agricultural Research,2015;10(36):3614–3624.
32. Pelizer LH, Danesi EDG, de O Rangel C, Sassano CE, Carvalho JCM, Sato S, et al. Influence of inoculum age and concentration in *Spirulina platensis* cultivation. Journal of food engineering,2003;56(4):371-375.
33. Rajasekaran C, Ajeesh CPM, Balaji S, Shalini M, Siva R, Das R, et al. Effect of Modified Zarrouk's Medium on Growth of Different *Spirulina* Strains. Walailak J. Sci. and Tech.,2016;13(1):67-75.
34. Raouf B, Kaushik BD, Prasanna R. Formulation of a low-cost medium for mass production of *Spirulina*. Biomass and bioenergy,2006;30(6):537-542.
35. Richmond A. Handbook of microalgal culture: Biotechnology and applied phycology. Blackwell Science, 2004.
36. Richmond J. A general method for constructing simultaneous confidence intervals. Journal of the American Statistical Association,1982;77(378):455-460.
37. Rippka R, Deruelles J, Waterbury JB, Herdman M, Stanier RY. Generic assignments, strain histories and properties of pure cultures of cyanobacteria. Microbiology,1979;111(1):1-61.
38. Salunke BK, Sawant SS, Lee SI, Kim BS. Microorganisms as efficient biosystem for the synthesis of metal nanoparticles: current scenario and future possibilities. World Journal of Microbiology and Biotechnology,2016;32(5):88.
39. Sánchez-Bayo A, Morales V, Rodríguez R, Vicente G, Bautista LF. Cultivation factors affecting microalgae production. Molecules,2020;25(12):2834.
40. Singh P, Dhar DW. Cyanobacteria and algal biotechnology for sustainable agriculture and environment. Journal of Applied Phycology,2020;32(1):1–15.
41. Soni RA, Sudhakar K, Rana RS. Comparative study on the growth performance of *Spirulina platensis* on modifying culture media. Energy reports,2019;5:327-336.
42. Torzillo G, Sacchi A, Materassi R, Richmond A. Effect of temperature on yield and night biomass loss in *Spirulina platensis* grown outdoors in tubular photobioreactors. Journal of Applied Phycology,1991;3(2):103-109.
43. Vonshak A. *Spirulina platensis (Arthrospira)*: Physiology, cell biology and biotechnology. Taylor & Francis, 1997.
44. Vonshak A, Laorawat S, Bunnag B, Tanticharoen M. The effect of light availability on the photosynthetic activity and productivity of outdoor cultures of *Arthrospira platensis (Spirulina)*. Journal of Applied Phycology,2014;26(3):1309-1315.
45. Vonshak A, Richmond A. Mass production of the blue-green alga *Spirulina*: an overview. Biomass,1988;15(4):233-247.
46. Vonshak A, Torzillo G. Environmental stress physiology. In: Richmond A (ed) Handbook of microalgal culture. Blackwell Publishing Ltd, Oxford, 2003, 57–82.
47. Zarrouk C. Contribution à l'étude d'une cyanophycée: Influence de divers facteurs physiques et chimiques sur la croissance et la photosynthèse de *Spirulina maxima*. University of Paris, 1966.