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Research Article

Growth, Productivity and Some Physico-chemical Factors of *Spirulina platensis* Cultivation as Influenced by Nutrients Change

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Abstract

Background and Objective: Microalga cells directly use nutrient elements, sunlight and CO₂ for their growth. Environmental factors such as temperature and pH affect metabolic process in microalga cells. The objective of this study was to evaluate the effect of nutrients change in the culture medium of *Spirulina platensis* (*S. platensis*) on its growth, productivity and some physico-chemical factors in order to define the optimal growth cultivation conditions. **Materials and Methods:** *Spirulina platensis* was grown in a 300 mL glass containers containing 100 mL nutrient medium at 30°C under 5000 lux illumination and 12:12 h light-dark cycle. *Spirulina platensis* cells were harvested after 42 days and analysed for dry weight (DW), specific growth rate (SGR) and productivity. Jourdan's medium was taken as control. For experiments, Jourdan's medium was enriched with iron sulphate (FeSO₄·7H₂O), sodium chloride (NaCl), magnesium sulphate (MgSO₄·7H₂O), urea (CO(NH₂)₂) and sodium bicarbonate (NaHCO₃) at different concentrations (0.01, 2.5, 0.1, 0.02 and 4 g L⁻¹, respectively). The temperature, pH and electrical conductivity (EC) were monitored for 42 days on daily basis. Averages of data were statistically analyzed by using analysis of variance (one-way ANOVA). Statistical differences between treatment means were established using the fisher LSD test at p<0.05. **Results:** The DW, SGR and productivity were positively influenced by the addition of FeSO₄·7H₂O, NaCl, MgSO₄·7H₂O, CO(NH₂)₂ and NaHCO₃ to Jourdan's medium at 0.01, 2.5, 0.1, 0.02 and 4 g L⁻¹, respectively. No significant change in pH values of all culture media of *S. platensis* throughout the study period was observed. The temperature of the culture was positively influenced by the addition of FeSO₄·7H₂O and NaCl to Jourdan's medium, while MgSO₄·7H₂O, CO(NH₂)₂ and NaHCO₃ lead to a decrease of the temperature from 31.66-25.90°C. Jourdan's medium supplied with 15 g L⁻¹ NaCl showed significantly (p<0.05) higher EC compared to all other treatments. The values of secchi depth were ranged from 2.06-3.02 cm when the media were enriched with NaCl, MgSO₄·7H₂O, CO(NH₂)₂ and NaHCO₃. **Conclusion:** The change of the nutrients in Jourdan's medium has the potential to produce a large scale biomass of *S. platensis* and could be suitable for its optimal growth culture conditions that could be beneficial for human's health.

Key words: *Spirulina platensis*, physico-chemical factors, artificial medium, nutrients amount, optimal growth

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Spirulina platensis is a cyanobacterium rich in proteins (60-70%), essential fatty acids, beta-carotene, vitamins and mineral elements¹. This alga (200-400 µm) lives in shallow water with a salinity ranging from 7-8% approximately and high alkalinity. The optimum pH for its survival is 9^{2,3}. Recent studies show that the bioavailability of nutrients in this alga was higher than that of dietary fiber^{4,5}. It may therefore be essential to use *S. platensis* as a supplement to protein, energy and as a means of avoiding malnutrition. Phycocyanin is extracted from *S. platensis*⁶. It is a blue pigment used in the cosmetic and food industries as the Lina blue. When highly purified, it has fluorescence properties that are used in immune diagnostic tests⁷. The performance of algae cultures in summer was particularly high in biomass (>300 mg DW L⁻¹) and yield (>30 mg DW L⁻¹ day⁻¹)⁸.

Mineral elements, sunlight and CO₂ are directly used by alga cells for their growth. Environmental factors such as temperature and pH are very essential in the micro algal cell metabolic processes⁹. During the cultivation of *S. platensis*, fertilizers have a direct impact on growth and photosynthesis¹⁰. To obtain biomass is a complicated issue during the large scale production of *S. platensis* since the process involves a large number of optimum conditions for the successful growth of the alga¹¹. Experiment performed on *Arthrospira* showed a decrease in the growth rate of microalga associated to the sources of nutrients¹². A deficiency in magnesium sulphate (MgSO₄.7H₂O) led to a breaking off cells in green microalgae resulting to abnormal large cell formation¹³. An increase in MgSO₄.7H₂O, in the culture medium resulted in higher cell number. A high alkalinity was required for the growth of *S. maxima* and high amount of NaHCO₃ was used to maintain high pH¹². The iron sulphate (FeSO₄.7H₂O) uptake was strictly required for phytoplankton development while the lack of FeSO₄.7H₂O in the culture medium retarded growth, reduced photosynthetic activity and chlorophyll content observed¹⁴. A number of green microalga species have been shown to be able to utilize carbonates such as Na₂CO₃ and NaHCO₃ for cell growth¹⁵. Nitrogen is often identified as limiting factor to algal biomass¹⁶. Nitrogen occurs in fresh water in numerous forms like dissolved nitrogen, amino acids, amines, urea, ammonium, nitrite and nitrate⁹. The best cellular growth was observed with 500 mg L⁻¹ of urea (CO(NH₂)₂) at a light intensity of 5600 lux, whereas, the highest concentration of chlorophyll in the biomass was observed with 500 mg L⁻¹ of CO(NH₂)₂ at a light intensity of 1400 lux¹⁷. Salinity (NaCl) was one of the conditions in which naturally thrives *S. platensis*. According to Iltis¹⁸, *S. platensis* has grown in salt water upto 60 g L⁻¹. The enrichment of a

medium with excessive NaCl was due to environmental contamination by foreign organisms but this medium should be diluted as soon as possible for optimum production¹⁹.

The growth of microscopic green microalgae appears to be more restricted by low temperature²⁰. The pH of a thriving culture was above 7.5 to limit contamination by foreign organisms^{21,19}. Beyond a pH value of 11.5, the cultivation of *S. platensis* was delayed²². The concentration of a culture can be assessed by the intensity of its colour. A secchi depth of 2-3 cm was an indication of appropriate culture production²³. This depth helps to manage the amount of heat in the medium. Values of secchi depth lower than 2 cm could indicate that it is necessary to dilute the culture, or harvest immediately since the only way to adjust these depth values were by increasing the temperature of the culture medium which in turn can destroy the filaments of *S. platensis*²³. In contrast, depths deeper than 3 cm help to decrease the temperature of the culture medium and thus delay the growth of *S. platensis*⁴.

Despite the use of organic fertilizers in order to increase fish production, or to purify the waste, monitoring interactions between inorganic nutrient sources, physico-chemical factors and intermediate biological organisms is relatively recent²⁰. Therefore, the objective of this study was to evaluate the effect of nutrients change in the culture medium of *S. platensis* on its growth, productivity and some physico-chemical factors in order to define the optimal growth cultivation conditions.

MATERIALS AND METHODS

Plant material and growth medium: Numerous studies showed that the bio-availability of nutrients in *S. platensis* is higher than that of dietary fiber^{4,5}. It may therefore, be essential to use *S. platensis* as a supplement to protein, energy and as a means of avoiding malnutrition in children. The present study was conducted in Faculty of Science, University of Douala, Cameroon, between March, 2013 to June, 2014. *Spirulina platensis* strain was obtained from Institute of Fisheries and Aquatic Sciences, Cameroon. *Spirulina platensis* was grown on Jourdan's medium consisting of (per L) 0.05 g CO(NH₂)₂, 0.12 g (NH₄)₂HPO₄, 2 g KNO₃, 0.15 g MgSO₄.7H₂O, 0.02 g CaCl₂, 0.02 g FeSO₄.7H₂O, 5 g NaCl and 8 g NaHCO₃. Jourdan's medium was taken as control. Moreover, for the 5 experimental groups, Jourdan's medium was enriched after autoclaving with FeSO₄.7H₂O at concentration of 0.01, 0.02 and 0.4 g L⁻¹, NaCl at 2.5, 10 and 15 g L⁻¹, MgSO₄.7H₂O at 0.1, 0.2 and 0.4 g L⁻¹, CO(NH₂)₂ at 0.02, 0.07 and 0.09 g L⁻¹ and NaHCO₃ at 4, 12 and 16 g L⁻¹. The microalga was grown in a 300 mL glass containers containing

100 mL nutrient medium at 30°C under illumination (5000 lux) and 12:12 h light-dark cycle and shaking (120 rpm).

Dry weight concentrations: For dry weight measurement homogenous suspension of *S. platensis* sample was filtered through Whatman No. 1 filter paper and oven dried at 75°C for 6 h. The dried filter paper containing *S. platensis* dry weight was cooled and weighted. The difference between the initial and final weight was taken as the dry weight of *S. platensis* biomass. The dry weight was expressed in terms of g L⁻¹.

Specific growth rate: The specific growth rate (μ) was calculated as follows²⁴:

$$\mu = \frac{\ln(X_2) - \ln(X_1)}{t_2 - t_1}$$

where, X₂ and X₁ represent the biomass concentrations at the times t₂ and t₁, respectively. The specific growth rate was expressed in terms of cell/day.

Productivity: The productivity (p) of *S. platensis* was determined according to the formula described by Jarisoa²⁵:

$$P = X_2 - X_1 / t_2 - t_1$$

where, X₂ and X₁ represent the biomass concentrations at the times t₂ and t₁, respectively. The productivity was expressed in terms of mg L⁻¹/day.

Densities of *S. platensis*: The secchi disk was used daily to measure the density of *S. platensis* in all containers. It is a ruler at the end of which is perpendicularly attached small white disk. This instrument was immersed in the culture to the point where the disk ceases to be visible. The depth of the disk was read from the scale.

Physico-chemical parameters: The temperature, pH and electrical conductivity were measured according to the methods described by Rodier *et al.*²⁶. The pH and temperature were measured by the HANNA multiparameter (HI 98127) and the electric conductivity using a HANNA conductivity meter (HI8733).

Data analysis: Data were presented in term of mean (\pm standard deviation). Data were analysed using statistical (version 9, Tulsa, OK, USA) and first subjected to one-way analysis of variance (ANOVA). Statistical differences between treatment means were established using the Fisher LSD test at $p < 0.05$.

RESULTS AND DISCUSSION

Dry weight concentrations, specific growth rate and yield of *S. platensis*: The effects of nutrients change on dry weight (DW), specific growth rate (SGR) and yield are depicted in Table 1. The obtained data showed that the Jourdan's medium enriched with iron sulphate (FeSO₄.7H₂O) at concentration of 0.01 g L⁻¹ lead to a significant ($p < 0.05$) increase in DW compared to control. On the contrary, the supply of Jourdan's

Table 1: Effect of nutrients change on Secchi depth, dry weight, specific growth rate, productivity and pH for 6 weeks of *S. platensis* cultivation

Nutrient sources	Amount (g L ⁻¹)	Secchi disk (cm)	Dry weight (g L ⁻¹)	Specific growth rate (cell/day)	Productivity (mg L ⁻¹ /day)	pH
FeSO ₄ .7H ₂ O	0.02 (SM)	1.82 ± 0.03 ^a	0.86 ± 0.07 ^d	0.06 ± 0.01 ^g	7.61 ± 0.24 ^j	10.55 ± 0.02 ^a
	0.01	1.94 ± 0.01 ⁱ	1.09 ± 0.08 ^b	0.07 ± 0.02 ^{fg}	79.65 ± 6.90 ^d	10.49 ± 0.03 ^a
	0.2	1.90 ± 0.02 ⁱ	0.68 ± 0.02 ^e	0.05 ± 0.01 ^g	2.07 ± 0.18 ^k	10.48 ± 0.01 ^a
	0.4	1.92 ± 0.03 ⁱ	0.60 ± 0.03 ^e	0.05 ± 0.01 ^g	1.03 ± 0.13 ^k	10.52 ± 0.02 ^a
NaCl	5 (SM)	2.65 ± 0.04 ^e	0.79 ± 0.05 ^d	0.08 ± 0.02 ^f	39.28 ± 2.81 ^g	10.24 ± 0.01 ^a
	2.5	2.06 ± 0.03 ^h	1.20 ± 0.05 ^a	0.09 ± 0.02 ^f	64.50 ± 5.31 ^e	10.31 ± 0.03 ^a
	10	2.68 ± 0.03 ^b	0.98 ± 0.04 ^{bc}	0.07 ± 0.01 ^{fg}	29.29 ± 1.68 ^{gh}	10.18 ± 0.04 ^b
	15	3.53 ± 0.06 ^b	0.11 ± 0.02 ⁱ	0.08 ± 0.02 ^h	7.20 ± 0.84 ^l	10.06 ± 0.02 ^b
MgSO ₄ .7H ₂ O	0.15 (SM)	2.10 ± 0.03 ^h	0.53 ± 0.05 ^f	0.46 ± 0.01 ^c	68.07 ± 6.16 ^e	9.93 ± 0.04 ^a
	0.1	2.50 ± 0.05 ^f	0.46 ± 0.03 ^f	0.48 ± 0.02 ^b	46.02 ± 4.60 ^{fg}	9.88 ± 0.02 ^{ab}
	0.2	2.35 ± 0.05 ^g	0.41 ± 0.02 ^{fg}	0.56 ± 0.06 ^b	25.80 ± 2.44 ^h	9.85 ± 0.03 ^{ab}
	0.4	2.46 ± 0.04 ^f	0.37 ± 0.04 ^g	0.52 ± 0.02 ^b	22.08 ± 2.82 ^h	10.01 ± 0.02 ^a
CO(NH ₂) ₂	0.05 (SM)	2.82 ± 0.04 ^d	0.72 ± 0.08 ^e	0.31 ± 0.03 ^{cd}	50.09 ± 5.23 ^f	9.80 ± 0.05 ^a
	0.02	2.47 ± 0.05 ^f	1.09 ± 0.10 ^b	0.45 ± 0.05 ^{bc}	90.10 ± 8.74 ^c	9.80 ± 0.03 ^a
	0.07	3.02 ± 0.08 ^c	0.67 ± 0.07 ^e	0.18 ± 0.01 ^e	45.65 ± 3.37 ^g	9.70 ± 0.04 ^b
	0.09	2.37 ± 0.06 ^e	0.37 ± 0.03 ^g	0.41 ± 0.03 ^c	56.04 ± 4.10 ^{ef}	9.40 ± 0.03 ^c
NaHCO ₃	8 (SM)	2.32 ± 0.02 ^g	0.87 ± 0.07 ^d	0.53 ± 0.03 ^{ab}	117.62 ± 6.25 ^b	9.79 ± 0.06 ^a
	4	2.02 ± 0.01 ^{hi}	1.03 ± 0.10 ^b	0.62 ± 0.05 ^a	168.07 ± 7.92 ^a	9.81 ± 0.07 ^a
	12	2.77 ± 0.03 ^d	0.62 ± 0.08 ^e	0.39 ± 0.02 ^c	60.41 ± 5.80 ^c	9.73 ± 0.02 ^{ab}
	16	3.63 ± 0.06 ^a	0.41 ± 0.03 ^{fg}	0.12 ± 0.01 ^f	40.08 ± 3.04 ^g	9.63 ± 0.04 ^b

Means (\pm SD, n = 5) with the same small letters in the same column are not significantly different at $p < 0.05$, FeSO₄.7H₂O: Iron sulphate, NaCl: Sodium chloride, MgSO₄.7H₂O: Magnesium sulphate, CO(NH₂)₂: urea, NaHCO₃: Sodium bicarbonate, SM: Jourdan's medium

medium with $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ at 0.2 and 0.4 g L⁻¹ showed significant ($p < 0.05$) reduction of DW. A significant ($p < 0.05$) increase was observed in the yield of *S. platensis* when $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ was added at a concentration of 0.01 g L⁻¹, while a decrease was found at 0.2 and 0.4 g L⁻¹ (Table 1). According to Borowitzka²⁷ and Guillou *et al.*²⁸, the DW and yield of microalga need low-dose of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ which is necessary for the production of chlorophyll. There was no significant difference in specific growth rate of *S. platensis* when the Jourdan's medium was supplied with different amounts of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ (Table 1). The enrichment of the Jourdan's medium by upto 10 g L⁻¹ NaCl had a positive effect on DW of *S. platensis* compared to the control (Table 1). In contrast, a significant ($p < 0.05$) reduction was observed when NaCl was added in Jourdan's medium at a concentration of 15 g L⁻¹ (Table 1). It is well known that cyanobacteria are able to adapt to the alkaline habitat, but high salinity could become limiting²⁹. According to Jourdan¹⁹, the enrichment of a medium with excessive NaCl may be due to environmental contamination by foreign organisms but this medium should be diluted as soon as possible for alga optimum production. There was no significant change in specific growth rate of *S. platensis* when the Jourdan's medium was enriched with different amounts of NaCl (Table 1). Jourdan's medium enriched with magnesium sulphate ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$) at different concentrations had a depressive effect on DW, SGR and yield of *S. platensis* (Table 1). Similarly, Vonshak and Tomaselli¹² in an experiment on *Arthrospira* reported a decrease in the DW, SGR and productivity of microalga due to the sources of nutrition. According to Finkle and Appleman¹³ and Rinanti *et al.*⁹, the only increase in $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ in the medium resulted in a high cell number but the process of multiplication requires a larger concentration of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ in the medium than the production of cell material. The addition of $\text{CO}(\text{NH}_2)_2$ in Jourdan's medium had a positive effect on the DW and yield of *S. platensis* compared to control (Table 1). These results could be explained by the absorption of nitrogen by the microalga. It is well known that that inorganic form of nitrogen (NH_4^+) was preferentially used by many algae. The NO_3^- and NO_2^- should be reduced prior to assimilation³⁰. There was a significant ($p < 0.05$) change in SGR of *S. platensis* when $\text{CO}(\text{NH}_2)_2$ was added at concentration of 0.07 g L⁻¹ in Jourdan's medium. Nutrient deficiency, especially nitrogen, may affect the cultivation of microalga in various ways. It seems that in nitrogen rich growth media, protein production was improved while carbohydrate synthesis was limited²⁸. In contrast, carbohydrate synthesis increases and protein production drops in nitrogen-deficient media³¹. It is depicted from Table 1 that NaHCO_3 has some influence on *S. platensis* cultivation. The Jourdan's medium enriched with

different amounts of NaHCO_3 leads to a significant ($p < 0.05$) increase in DW, SGR and yield of *S. platensis* compared to control (Table 1). The highest increase (15.5, 14.5 and 30.0%) of DW, SGR and yield of *S. platensis* was recorded under NaHCO_3 treatment at 4 g L⁻¹, respectively (Table 1). Similarly, Juneja *et al.*³² found that carbon dioxide (CO_2) reacted with the carbonates and his consumption by microalga during photosynthesis is result in an increase in pH due to the release of the OH^- which affects the ionization of certain ions. Thus, the NH_4^+ form becomes more abundant when the pH rises²⁰. High alkalinity was required for the growth of *S. maxima* and a higher amount of NaHCO_3 was used to maintain high pH¹². In addition, CO_2 must be added into the culture medium to maintain optimum pH due to the fast growth of the biomass at elevated temperature²⁴. A number of green alga species have a high extracellular carboanhydrase activity which was responsible for the conversion of carbonate to free CO_2 to facilitate CO_2 assimilation¹⁵.

Densities of *S. platensis*: The values of secchi disk used to measure the densities of *S. platensis* in all containers showed a significant ($p < 0.05$) reduction when $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, NaCl and NaHCO_3 were supplied at low levels 0.01, 2.5 and 4 g L⁻¹, respectively (Table 1). On the contrary, the application of these nutrients at higher levels 0.4, 15 and 16 g L⁻¹, respectively lead to an increase of the values of secchi depth during 6 weeks of *S. spirulina* cultivation compared to the control (Table 1). The highest value of the secchi depth (3.63 cm) was recorded when NaHCO_3 were added in Jourdan's medium at 16 g L⁻¹ (Table 1). According to Cruchots⁴, the value of secchi depth above 3.0 cm helps to decrease the amount of heat in the medium and thus delayed the growth of *S. platensis*. On the contrary, the addition of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ influenced positively the secchi depth after 6 weeks of *S. platensis* cultivation (Table 1). The highest value (2.50 cm) was found at 0.1 g L⁻¹ when the artificial medium was enriched with $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ (Table 1). The addition of $\text{CO}(\text{NH}_2)_2$ had a positive effect on secchi depth compared to the control (Table 1). In the present study, the values of secchi depth varied and the magnitude of variation relied on the levels and sources of nutrients used. These values ranged from 2.06-3.02 cm when the media were enriched with NaCl, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CO}(\text{NH}_2)_2$ and NaHCO_3 except for NaHCO_3 at 16 g L⁻¹. Similarly, Falquet and Hurni⁵ reported that the values of 2-3 cm of secchi depth correspond to a ready culture production of *S. platensis* and these depths may help to manage the amount of heat in the medium. The values of secchi depth were less than 2 cm when $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ was supplied at different concentrations. According to Falquet²³, the values lower than 2 cm could indicate that it is necessary to dilute the culture medium or harvest

immediately because the only means of adjusting these depth values were by increasing the amount of heat in the culture medium which subsequently destroy the *S. Platensis* filaments.

pH values: The addition of nutrients did not change too much the pH value of culture medium during the study period (Table 1). The pH of culture medium became more basic as compared to control. The pH of a thriving culture is between 9 and 10.5 reflecting the vitality of microalga cultivation^{19,20,33}. The pH should be above 7.5 to limit contamination by foreign organisms. When beyond 11.5, the cultivation of *S. platensis* was delayed¹⁹. Similar results were reported by

Seshadri *et al.*²². According to Vonshak and Tomaselli¹², these results indicated very poor growth in respective nutrient concentrations. *S. platensis* requires relatively high pH values between 9.5 and 9.8 which inhibit the contamination of most microalgae in the artificial medium¹¹. Maintaining such a high pH prevents contamination from other harmful bacteria such as *Escherichia coli* and intestinal parasites such as *Schistosomiasis* and *Giardiasis*³⁴. In this respect, high amounts of NaHCO₃ must always be present in the culture medium to sustain the high pH and prevent fluctuations²⁴.

Temperature of the culture medium: The temperature of the culture fluctuated from 25.9-31.7°C (Fig. 1) in accordance with

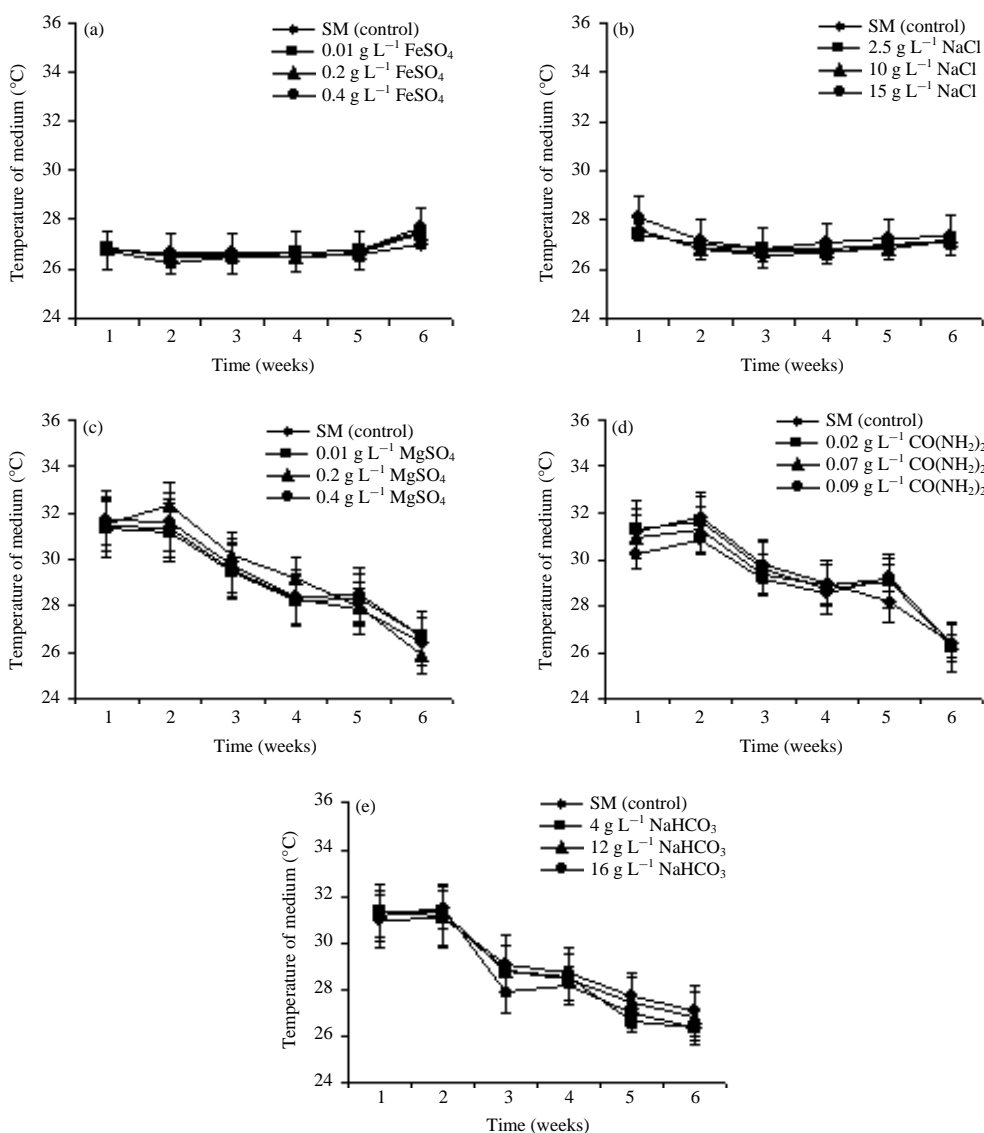


Fig. 1 (a-e): Effect of nutrients change on temperature for 6 weeks of *S. platensis*, (a) FeSO₄.7H₂O: iron sulphate, (b) NaCl: Sodium chloride, (c) MgSO₄.7H₂O: Magnesium sulphate, (d) CO(NH₂)₂: Urea and (e) NaHCO₃: Sodium bicarbonate. Mean ± SD and n = 5. Bars indicate standard deviation

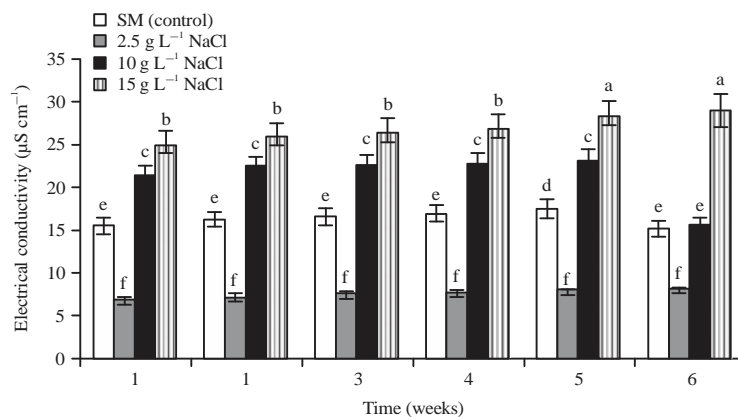


Fig. 2: Effect of NaCl concentrations on electrical conductivity for 6 weeks of *S. platensis*. Means (\pm SD, n = 5) with the same small letters are not significantly different at $p < 0.05$. Bars indicate standard deviation

results from other authors^{27,35}. The addition of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ decreased the temperature of the culture medium in the first 2 weeks and later stabilized for 4 weeks and finally increased at the end of the experiment (Fig. 1a). The temperature of the Jourdan's medium supplied with $0.4 \text{ g L}^{-1} \text{ FeSO}_4 \cdot 7\text{H}_2\text{O}$ was highest compared to all other treatments (Fig. 1a). It was observed that an increase in temperature lead to a marked decrease in protein content while carbohydrate synthesis was stimulated in *S. platensis*³⁶. According to Renaud *et al.*³⁷, some Australian microalgae had significantly lower percentages of protein when cells were grown at temperatures above 27°C , but there was no consistent trend in the percentages of carbohydrate. The temperature was affected by NaCl concentrations (Fig. 1b). The addition of NaCl decreased the temperature in the first 3 weeks and increased in the last 3 weeks of the experiment (Fig. 1b). According to Borowitzka²⁷, higher temperatures promote the growth of alga compared to lower temperatures. According to Richmond *et al.*³⁸, the temperature of the medium exerted a predominant effect on *S. platensis* production. The temperature of the culture medium was notably decreased in response to $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ amendment after 6 weeks of *S. platensis* cultivation (Fig. 1c). Jourdan's medium supplied with $0.4 \text{ g L}^{-1} \text{ MgSO}_4 \cdot 7\text{H}_2\text{O}$ had the highest temperature compared to all other treatments while the lowest was found at $0.2 \text{ g L}^{-1} \text{ MgSO}_4 \cdot 7\text{H}_2\text{O}$ (Fig. 1c). Similarly, Dabbadie²⁰ found that the growth of microscopic green alga appears to be more restricted by low temperature. The addition of $\text{CO}(\text{NH}_2)_2$ and NaHCO_3 , respectively decreased the temperature of the culture medium after 6 weeks of *S. platensis* cultivation (Fig. 1d, e) compared to control. Temperature is one of the most crucial factors affecting biomass accumulation and lipid

production by alga cells since it is the focal point in all enzymological reactions and physiological functioning of the cells³⁵.

Electrical conductivity of the culture medium: The results showed a significant ($p < 0.05$) difference between the amount of NaCl added and the electrical conductivity (EC) in *S. platensis* culture medium (Fig. 2). The EC was highest ($29.15 \mu\text{S cm}^{-1}$) when NaCl was added at 15 g L^{-1} and lowest ($7.79 \mu\text{S cm}^{-1}$) when the medium was enriched with 2.5 g L^{-1} during the 5 weeks of cultivation of *S. platensis* compared to control. The EC is an important parameter to measure the utilization of inorganic materials in the medium by alga cells. According to Mutanda *et al.*³⁵, the phenomenon observed is explained by bioavailability in chemical species in the BG-11 medium whose uptake by the microalga cells led to a decrease in EC in the microalga culture. Values of EC ranging from $7.65\text{-}29.15 \mu\text{S cm}^{-1}$ are too low and could reveal the low mineralization in the aqueous *S. platensis* suspension²⁶.

CONCLUSION

The present study shows that the dry weight, specific growth rate and yield of *S. platensis* were positively influenced by the addition of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, NaCl, $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CO}(\text{NH}_2)_2$ and NaHCO_3 to Jourdan's medium at 0.01 , 2.5 , 0.1 , 0.02 and 4 g L^{-1} , respectively. This suggests that the change of the nutrients in Jourdan's medium has the potential to produce a large scale biomass of *S. platensis* and could be suitable for its optimal growth culture conditions. The addition of different amounts of those nutrients did not significantly change the pH values ($9.40\text{-}10.55$). The supply of Jourdan's

medium with $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CO}(\text{NH}_2)_2$ and NaHCO_3 showed negative effect on temperature while different amounts of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ and NaCl showed positive impact. The electrical conductivity was closely associated to salinity. The values in the aqueous microalga suspension were higher ($29.15 \mu\text{S cm}^{-1}$) when NaCl was added at 15 g L^{-1} and lower ($7.89 \mu\text{S cm}^{-1}$) in the medium enriched with 2.5 g L^{-1} . The values of secchi depth ranged from 2.06-3.02 cm when the culture media were enriched with NaCl , $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{CO}(\text{NH}_2)_2$ and NaHCO_3 except for NaHCO_3 at 16 g L^{-1} .

SIGNIFICANCE STATEMENTS

This study discovers that the change of amounts of the nutrients in artificial medium has the potential to produce a large scale biomass of *Spirulina platensis* and could be suitable for its optimal growth cultivation conditions that could be beneficial for human's health. This study will help the researcher to uncover the critical areas of the improvement of nutritional situation of malnourished children in the development country that many researchers were not able to explore. Thus a new theory on a food way of improving nutritional status may be arrived at.

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