ENHANCING BIOMASS PRODUCTION BY THE OPTIMIZATION OF PROCESS PARAMETERS OF MICRO ALGAE (ARTHROSPIRA PLATENSIS) IN CONICAL ANNULAR PHOTOBIOREACTOR USING RESPONSE SURFACE METHODOLOGY

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ABSTRACT

In this experimental study, a novel cultivation mode in a conical annular photobioreactor (CAPBR) was proposed to enhance microalgae biomass production of Arthrospira platensis. Three operating factors under varying conditions of initial inoculum concentration (0.25–1.25 g.L−1), light intensity (6–180 µmol.s−1.m−2) and surface/volume (S/V) ratios (94–112 m−1) were optimized. Box-Behnken design combining with response surface methodology was used to evaluate the effect of operating parameters on response variables (Volumetric productivity, Growth rate). Optimal parameters of the chosen variable were found by considering the quadratic model, as well as by evaluating the desirability function. The obtained results showed that the light intensity and S/V ratio were the most significant factors conditioning volumetric productivity and growth rate. Moreover, an important interaction between light intensity and S/V ratio (P < 0.05) on volumetric productivity and growth rate was reported.

The maximum volumetric productivity and growth rate of 542 mgL−1.day−1 and 0.785 day−1 respectively was obtained at 180 µmol.s−1.m−2, with inoculum concentration of 0.351 g.L−1 and S/V ratio of 112 m−1. According to ANOVA analysis, this model reasonably predicts match the experimental data.

Keywords: Photobioreactor, Biomass productivity, Arthrospira platensis, optimization, Box-Behnken design.

Introduction

The industrial production of phototropic microorganisms is gaining momentum in different fields particularly in as food and pharmaceutical industries. Photosynthetic microorganisms present a most attractive energy source because it is renewable and without greenhouse effect. Among the photosynthetic microorganisms of commercial interest, Arthrospira platensis is one of the most interesting species. It is unbranched spiral filamentous with 200 to 300 µm in length and 5 to 10 µm in width (Benelhadj et al., 2016). They are filamentous cyanobacteria that are commonly found in the marine environment under highly alkaline and brackish living conditions, their commercial applications are prospective, widely used for the production of organic foods, cosmetics, food additives, pigments with important antioxidant molecules and anti-inflammatory properties (Pedrosa et al., 2011; Larrosa et al., 2018; Hadizadeh et al., 2019; Manirafasha et al., 2018; and Chaiklahané et al., 2022). Chlorella and Arthrospira are the most prominent species in the microalgae market due to their primary uses as human nutrition (Moroch-Jâcome et al., 2016). In 1974, the word Food Conference of United Nations declared Arthrospira as “the best for tomorrow” (UNWFC, 1974).

Arthrospira platensis is a blue-green alga rich in balanced proteins and great source of various nutraceuticals, has a particular chemical composition with 55 to 70% of proteins, 15 to 25% of carbohydrates and 4 to 7% of lipids (Ljubic et al., 2018). It has been successfully produced in open raceway ponds for commercial use (Lu et al., 2011). Therefore, special strategies have to be introduced to increase A. platensis productivity as different factors could impact biomass yield including, the mode of the operation, light intensity, cell concentration and reactor design (Chaiklahané et al., 2022; da Silva et al., 2016; Li et al., 2021; Bezerra et al., 2011).

According to previous studies, Arthrospira platensis can be cultivated in open ponds and closed photobioreactor, mainly with tubular configuration (Ferreira et al., 2012; Morocho-Jâcome et al., 2012; Xie et al., 2015). In open and closed cultures, various parameters greatly influence the growth of Spirulina platensis including pH, light intensity, inoculum level and culture contamination (Xie et al., 2015; Vonshak et al., 1982; Richmond, 2004; Danesi et al., 2011).

The optimal bioreactors have to present a high specific surface with high Surface/volume (S/V) ratio for efficient transfer of light energy. The current culture systems include batch operated shallow open ponds (15-30 cm of depth) (Belay, 1997), tubular photobioreactors (Carlozzi et al., 2000; Molina et al., 2001) and flat plat photobioreactors (Sforzal et al., 2014).
In the study of process optimization, generally, the “one parameter at time” approach is widely used, where only one parameter varied while the others are held constant. This method can be used as a rough assess of optimal levels (Norfarina et al., 2017).

This strategy necessitates a long time and costs a lot of money because there could be numerous experiments required (Adinarayana et al., 2002; Androga et al., 2014). In this respect, statistical experimental designs such as factorial design and response surface method are interesting alternatives. Response surface methodology (RSM) is a series of statistical methods for the experiment design, the creation of models, the estimation of the effect of variables and the search for optimal conditions desirable response factors (Bas et al., 2007; Haddar et al., 2010; Sathendra et al., 2019; Montgomery et al., 2012). The main advantage of RSM is the limited required experimental studies to test different parameters and to take into account their interactions (Ojha and Das, 2018). Several research works on the use of experimental design to optimize variables for biomass production are reported in literature including: biomass production by open raceway pond (Radmann et al. 2007; Junique et al., 2021), close photobioreactors (Carlozzi et al., 2003; Ho et al., 2012), phototrophic and mixotrophic cultures growth (Chojnacka et al., 2003; Verma et al., 2020).

In phototrophic growth through batch culture always ends with a stationary phase when environmental factors become unfavorable such as depletion of nutritional source (C/N), proton potential (pH), and viscosity. Another factor is added to the photobioreactor system which is the availability of light by the effect of light attenuation at the depth of the culture tank particularly for high cell density (Richmond, 2004).

In this context, we are looking for the combined effect of the specific surface ratio (S/V) with culture parameters that have positive interactions to minimize the limiting effect of attenuation and improve system performance with the goal of increasing productivity and maximizing final biomass concentration. Relatively, few data are published in this respect, and it remains interesting to investigate the interaction of surface area with other production parameters including light strength and inoculum concentrations.

The contribution of this work is the use of a new conical-annular PBR design with a high S/V ratio and the coupling of the specific surface area ratio (S/V) of the PBR with the light intensity and the inoculum concentration. This study was conducted using the design experiment approach. Regression models of biomass productivity and growth rate to the independent variables (initial biomass concentration, light intensity and ratio S/V of PBR) and their interaction were developed and optimized to get the best results.

Material and Methods

Bacteria and culture media

*Arthrospira platensis* used in this research was collected from the Laboratory of Biotechnology and Process Engineering, ENP Elharrach, Algerie. For the production of spirulina culture, the microalgae were seeded on synthetic Zarrouk medium (Zarrouk, 1966).

Experimental procedure

As shown in the figure, 1, the photobioreactor used in this research consisted of a conical glass vessel and annular structure with 60° inclination. This tilt allows the light to be projected perpendicularly on the PBR surface and to reduce radiation losses by bending. In addition, this configuration provides a 360° exposure surface and a short light path, allowing for light homogenization in the culture's volume.

This reactor significantly improved the production of H₂ by *S. platensis* compared to conical and cylindrical PBR (Ainas et al., 2017). The CAPBR is characterized by an external cone (Ec), an internal cone (Ic) and an annular chamber of small thickness (17-25 mm) with a high surface/volume(S/V) ratio (greater than 90 m⁻³). The top opening of the annular chamber is capped with an outlet to limit the entry of air borne contaminants, which creates a low positive pressure inside the reactor that further reduces contamination risks and agitation culture. It is provided by air sparging filtered using an air pump. The CAPBR was kept in a temperature-controlled room (to maintain the temperature at 30°C ± 2, illumination was provided by Osram 40 W (36 µmol m⁻² s⁻¹) daylight type fluorescent lamp under 12h photoperiod.

In order to evaluate the influence of initial inoculum concentration, light intensity and S/V ratio on volumetric productivity (Pᵥ) and growth rate (µᵥmax). three PBRs of different capacities (Table 1) were tested.

Measurements and analytical methods

*A. Platensis* concentration was calculated by measuring the optical density (OD) at 618 nm by spectrophotometer (model Shimadzu UV -1201, Tokyo, Japan) and expressed in gram of dried biomass per liter of medium (g.L⁻¹). At 618 nm a DO 618 nm of 1.0 corresponded to 0.799 g DW/L.

The biomass production performance was quantified by growth rates and volumetric productivity. The growth rate µᵥmax (estimated from the linear phase during exponential bacterial growth) is calculated by equation (1).

\[
µᵥmax = \frac{\ln X_2 - \ln X_1}{(t_2 - t_1) \ln 2}\frac{1}{\ln X_2 X_1} \quad ... (1)
\]

Where X₁ and X₂ correspond to the biomass concentration (g.L⁻¹) at time t₁ and t₂ (days), respectively.

The volumetric productivity (Pᵥ) was defined by the equation (2).

\[
Pᵥ = \frac{C_b - C_{b0}}{t - t_0} \quad ... (2)
\]

Where C_b and C_{b0} are the biomass concentration at time t and the starting time t₀, respectively.

The chlorophyll-a content in microalgae cells was estimated using a spectrophotometric method at 665, 645 and 630 nm. Chlorophyll contents in mg/g became calculated the usage of the following equation (Ruiz et al. 2015).

\[
\text{Chlorophyll-a (mg/g)} = 11.6(A_{665})-1.31(A_{645})-0.14(A_{630}) \quad (3)
\]

Experimental design
Response surface methodology is an important technique for optimizing a multivariate system. It is an empirical statistical modeling used for multiple regression using quantitative data derived from properly designed experiment (Rao et al., 2000).

The RSM was used in combination with Box-Behnken design through Design Expert software trial version 13 to examine the effect of three independent variables on response functions and to evaluate the optimum conditions for maximizing biomass production by *Arthopyra platensis* batch cultures. The optimization method includes observing the response of the statistically modeled combination, evaluation the coefficients by fitting the experimental data to the response function. Predicting the response of the fitted model and testing the model’s suitability (Wang et al., 2016).

The three independent factors in this research were the initial inoculum concentration ($X_1$), light intensity ($X_2$) and $S/V$ ratio ($X_3$). The response functions were the volumetric productivity ($P_V$) and growth rate ($\mu_{\text{max}}$). The range and the levels of the factors are given in Table 2. In the development of the statistical model, the relation between the coded values and actual values is described by Eq (4).

$$X_i = \frac{x_i - x_{i0}}{\Delta x_i}$$  

... (4)

Where $x_i$ and $X_i$ are coded and uncoded values of the $i$th independent variable: $X_{i0}$ is the uncoded values of the $i$th independent variable at the center point, and $\Delta x_i$ is the step change value between low level (-1) and high level (+1).

The quadratic model was also used to correlate the relation between independent variables and dependent variables, estimated with the following Eq (5)

$$Y = \beta_0 + \sum_{i=1}^{k} \beta_i X_i + \sum_{i=1}^{k} \sum_{j=1}^{k} \beta_{ij} X_i X_j + \varepsilon$$  

... (5)

Where $Y$ is the predicted response, $\beta_0$ intercept the term, $\beta_i$ the coefficient of linear effect, $\beta_{ii}$ the coefficient of squared effect, $\beta_{ij}$ the coefficient of interaction effect and $\varepsilon$ the random error.

$X_i$ and $X_j$ are the input n variables which influence the response of variable $Y$.

The Analysis of variance (ANOVA) was used to determine the relationship between the factors and the response variable. The quality of fit of the predictive model was expressed by $R^2$ and its statistical significance was tested with the F- test. Plots of the contour (2D) and response surface (3D) were used to investigate the influence of variable interaction over response (Behera et al., 2020).

**Results and Discussion**

The application of RSM provides an analytical relationship between the response function and the independent factors. Model terms were evaluated by the P-value (productivity) with confidence level of 95%. The contours plots were obtained according to the effects of the three factors (initial inoculum concentration, light intensity and surface/volume ratio) at three levels.

Table 3 present the experimental biomass production results from a three-factor and three-level factorial design of experiment.

**Fitting of quadratic mathematical model and analysis of variance**

The results of the study were fitted by an empirical relation represented by a $2^{nd}$ polynomial equation with interaction terms. Based on findings of the experimental design (Table 3), the regression models with codes factors for describing volumetric productivity ($P_V$) and growth rate ($\mu_{\text{max}}$) are seen in Eqs. (6) and (7) respectively.

$$P_V = 0.2281x_{1} - 0.0175x_2 - 0.0196x_3 + 0.1016x_1x_2 + 0.0789x_1x_3 - 0.0651$$  

... (6)

$$\mu_{\text{max}} = 0.035x_1 + 0.073x_2 + 0.079x_3 + 0.110x_1x_2 - 0.019x_1x_3 + 0.079x_2x_3$$  

... (7)

An analysis of variance was used to assess the suitability of the fitting of the second-order mathematical function to the experimental results. Table 4, displays the ANOVA analyses for volumetric productivity and growth rate. This analysis indicated that the $P_V$ model was significant, as shown by the F-value of 20.89, which had a p-value of 0.19 percent. The P-values quantify the statistical significance of each variable, low P-values indicated that the variables in the model are statistically significant (Liu et al., 2003).Moreover, the ANOVA result for growth rate model shows F-value of 14.53 expressing a significant effect on the response with $p = 0.44\%$ (Table 4). In addition, quadratic model statistics show high reliability in the estimation of volumetric productivity and growth rate ($R^2$ of 0.974 and $R^2$ of 0.963, respectively). A high correlation coefficient ensures a satisfactory adaptation of the quadratic model to the experimental results. The P-values (as seen in Table 5) were used to evaluate the significant and effective terms of the established models. The lower P value (below 0.05 at 95 % significance) for a model term. The P value greater than 5% is negligible as it indicates that the corresponding coefficient in the model is insignificant.

Diagnostic plots such as predicted versus actual value plots and standard probability plots of the studentized residual confirm the suitability of the chosen model as a reliable approximation of the real system. To evaluate a model’s suitability, these plots are used.

Fig. 2 (a and b) confirms that the predicted response values from the models are well aligned with the observed values; the data points are distributed relatively close to the single direction ($y = x$). These graphs show an acceptable agreement between the actual data and the data derived from the models. Data were also examined to confirm the residues' normality. Figure 3(a-b) presents the normal probability graph for these residues. This plot's data points are quite closely to a straight line, may be inferred that the data are uniformly distributed.
Evaluation of variables effect on the volumetric productivity and growth rate

Using RSM by the combined effect of independent variables offers prediction more than the conventional methods. In order to investigate the effect of each factor and their interactions on the responses, 3D surface response plots and their corresponding contour plots for the predicted response were developed. This plot, which is based on the model mathematical equation to analyze the change response surface, can facilitate the truthful examination of the effects of the experimental factors on the responses (Yetilmezsoy et al., 2009). In a 3D surface plot, a graphical representation of the quadratic model showing the impact of each variable on the obtained response. These graphs assist in explaining the effect of two unrelated factor on the response while retaining the third factor constant.

The contour plots offer the opportunity to determine the type of interactions that exist between variables. The spherical shape of the response surface suggests that there is negligible interaction. On the other hand, the elliptical shape of the contour plots indicates that the interaction between two variables is important (Muralidhar et al., 2001).

The surface plots for the volumetric productivity (Pv) and growth rate (µ_max) as a function of inoculums concentration, light intensity and S/V ratio, as formulated by equation 7 and 8 is presented in Figs. 4 and 5.

The combined effect of inoculums concentration and light intensity on volumetric productivity and growth rate is shown in Figs 4(a) and 5(a). The effect of the light intensity was investigated in the range of 6-180 µmol m^(-2) s^(-1). According to the results shown in Table 5, the effect of light intensity on the volumetric productivity and growth rate was significant (P value < 0.05).

While keeping the S/V ratio constant, it was deduced from the P-values of 0.4266 and 0.4975 there was no significant interaction between inoculum concentration and light intensity (Table 5).

The monitoring of both productivity and growth rate under the effect of the two tested variables reveals a higher volumetric productivity and growth rate of 0.51 gL^(-1) and 0.558 day^(-1) respectively with a lower inoculum and a higher light intensity and growth rate when the light intensity decreases from 180 to 6 µmol m^(-2) s^(-1). Therefore, it appearing that, in order to stabilize volumetric productivity at these ideal levels, a light intensity of 180 µmol m^(-2) s^(-1) and an initial inoculums concentration of 0.25 gL^(-1) must be introduced.

The combined effect of the two variables, light intensity and specific surface of PBR (S/V) on volumetric productivity and growth rate is illustrated in Figs. 4b and 5b an increase of both volumetric productivity and growth rate are observed with the increase of the two variables (I, S/V) at alight intensity of 6 µmol m^(-2) s^(-1), the volumetric productivity are generally less than 0.157 gL^(-1) day^(-1); however, the maximum volumetric productivity (0.51 gL^(-1) day^(-1)) was observed at 180 µmol m^(-2) s^(-1) and S/V 112 m^(-3) (Fig. 4b).

The 3D plot (Fig. 4b, 5b) also reveals that the volumetric productivity and growth rate was more sensitive to the change of the two variables, it presents a steeper slope between the two axes (axis of light intensity and axis of the S/V ratio). As shown in the Fig. 4(b), the contour plots are elliptical shape for volumetric productivity response, showing that interactive effect are no negligible. This was verified by the results of ANOVA in Table 5, in which the P-value is less than 0.05 for X_1X_3. On the other hand, Fig. 5(b) also shows a significant interaction (P value < 0.05) between light intensity and S/V ration on the growth rate.

The presence of such a linear response can be interpreted by the independence of the volumetric productivity and growth rate responses to the simultaneous variations of two studied parameters. The increase in light intensity and S/V ratio leads to increased biomass productivity which is essentially conditioned by the efficient conversion of light energy (Vonskak et al., 2014).

Figures 4(c) and 5(c) shows the effect of initial inoculum concentration and the specific surface area (S/V ratio) on volumetric productivity and growth rate at constant light intensity of 93 µmol m^(-2) s^(-1). The results showed the best value in terms of PV and µ_max (0.367 gL^(-1) day^(-1), 0.387 day^(-1) respectively) are obtained at a high S/V ratio and a less culture density.

According to table 5, the P-value (0.6420 and 0.7898) imply that the interactive (X_1*X_3) is insignificant terms for volumetric productivity and growth rate.

Application of photobioreactor with reduced light path (higher S/V ratio) allows more effective use of light and achieves the highest productivity (Gupta et al. 2015).

A low productivity of culture can be explained by the effect of self-shading by microalgae filaments when the biomass concentration in the broth becomes too dense leading to poor growth (Grima et al., 1996; Richmond, 2004).

Under these conditions, Ogbonna and Tanaka (2000) proposed that this is related to the direction of the phototrophic cell metabolism towards heterotrophy when the illumination is limited.

However, the effect of surface/volume ratio and light intensity on maximum biomass production was investigated. As shown in Fig. 6, a maximum biomass concentration of A. Platensis was reached at 4,548 g/L after 7 days and then stopping growth. Bezerra et al. (2011) reported that as the capacity of light to penetrate the culture determines the cell concentration in the saturation region. This behavior can be attributed to photo-saturation or autchading of the culture (Bezerra et al. 2016). In addition, it is widely recognized that illumination is attenuated exponentially upon entry into the culture, trying to impose strict limits on cell density culture (Ferreira, 2016).

In order to investigate the influence of light intensity and S/V ratio on chlorophyll content during A. platensis cultivation, the two vital parameters were maintained 6, 93 and 180 µmol m^(-2) s^(-1) and 94, 103 and 112 m^(-3).

Figure 7, shows chlorophyll-a content measurements performed on A. platensis cells grown under different light intensity and S/V ratio.

The result in figure 7, show a proportionality between the chlorophyll-a concentration obtained and the light intensity applied with a maximum chlorophyll concentration of 5.792 mg/g for the CAPBR with à low S/V ration of 94 m^(-3) and an intensity of 180 µmol m^(-2) s^(-1). However, in shallow CAPBR of 103; 112 m^(-3), the chlorophyll content increased...
linearly with light intensity ranging from 6 to 93 μmol m⁻² s⁻¹ with maximum chlorophyll-a values of 4.28; 3.218 mg/g for two dimensions 103 and 112 m⁻¹ respectively (Fig.6). On the other hand, at high intensity in the order of 180 μmol m⁻² s⁻¹, the chlorophyll-a concentration obtained from the two dimensions 103 and 112 m⁻¹ were 2.98 and 2.704 mg/g, respectively. These last results showed that there is a reduction of 16 and 30% respectively compared to the maximum values obtained at the intensity of 93 μmol m⁻² s⁻¹ (Fig. 7).

For algae, modification in their photosynthesis process to light have an adaptive significance with the aid of maximizing their capability increase. At light limitation, microalgae operate less photosynthesis and produce less energy. To compensate for this lack their chlorophyll content increase a two to ten times, these adjustments known as light-shade adaptations (Candioti et al., 2014). However, at high level of light intensity (180 μmol m⁻² s⁻¹), a reduction in concentration of chlorophyll likely due to photo inhibition effects in this irradiance or may be due to an acclimatization response by reducing Chlorophyll-a content to decrease light harvesting ability and an accumulation of other pigments like phycocyanin, carotenoids which have antioxidant activity in these conditions (Sforza et al., 2014).

The outcomes of this work were compared with other research published in the literature on the optimization of biomass productivity and growth rate. Table 7, summarizes some experimental studies, combining various environmental parameters such as light intensity and S/V ratio have an important impact on biomass production. In addition, the Pb and µmax obtained from this study is consistent with past research.

**Optimization by response surface methodology**

In order to find the best process condition, numerical optimization was conducted using the RSM. The desirability function (D) is used in Design-Expert software to select the ideal parameters of predicted volumetric productivity and growth rate. Desirability typically ranges from 0 to 1. A greater value signifies that the response is more desirable, while a zero value signifies that the response is completely undesirable (El-Mekkaoui et al., 2020).

The operational factors (inoculums concentration, light intensity and S/V ratio) were kept in range while the volumetric productivity and growth rate were set at maximum. Table 6 shows the optimal conditions with the estimated and experimental of the volumetric productivity and Growth rate. These optimal variables were chosen according to the highest desirability of 0.98.

At the suggested operating conditions of 0.35 gL⁻¹, 180 m² s⁻¹ and 112 m⁻¹ initial inoculums concentration, light intensity and S/V ration respectively, there was no discernible difference between the experimental and predicted data of the two models in terms of productivity and growth rate. Therefore, the predictive model was found to be suitable for predicting and optimizing the microalgae cultivation mode.

**Conclusion**

In this research, an annular conical photobioreactor was used to enhance biomass production from cyanobacterium *A. platensis* by the Box-Behnken (BBD) design. The impact of inoculums concentration, light intensity and S/V ratio have been studied by RSM in attempt to optimize growth conditions. The correlation between the independents variables and the biomass production was obtained by a 2nd polynomial function. According to the results, light intensity and S/V ratio were the most important factors affecting volumetric productivity and growth rate, while inoculums concentration had minor effect on growth rate. However, significant interaction was observed between light intensity and S/V Ratio (P value < 0.05) on the volumetric productivity. Nevertheless, a significant interaction between light intensity and S/V ratio was found to affect volumetric productivity (P value < 0.05. This study shows that *A. platensis* can be cultivated in CAPBRs with the maximum volumetric productivity, growth rate and chlorophyll content (0.542 g L⁻¹ day⁻¹, 0.785 day⁻¹, 2.704 mgg⁻¹) respectively, when the light intensity and S/V ratio are suitable.

The predictive model of RSM is found to be able to better predict the maximum biomass production under the imposed conditions.
Enhancing biomass production by the optimization of process parameters of micro algae (*Arthrospira platensis*) in conical annular photobioreactor using response surface methodology

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<th>X&lt;sub&gt;2&lt;/sub&gt;: Light intensity (µmol m&lt;sup&gt;-2&lt;/sup&gt; s&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>X&lt;sub&gt;3&lt;/sub&gt;: S/V ratio (m&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>Volumetric productivity (g L&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;)</th>
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**Table 4**: Analysis of variance (ANOVA) for the volumetric productivity and growth rate model.

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<td>0.0017</td>
<td>100.08</td>
<td>0.0099</td>
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<td>Pure error</td>
<td>2</td>
<td>0.0000</td>
<td>0.0000</td>
<td></td>
<td></td>
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<tr>
<td>Total</td>
<td>14</td>
<td>0.2022</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>R&lt;sup&gt;2&lt;/sup&gt;</td>
<td></td>
<td>0.974</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Adj-R&lt;sup&gt;2&lt;/sup&gt;</td>
<td></td>
<td>0.9274</td>
<td></td>
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**Table 5**: Coefficients from the RSM optimization for the volumetric productivity and Growth rate model.

<table>
<thead>
<tr>
<th>Term</th>
<th>Volumetric productivity, g.L&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;</th>
<th>Growth rate, day&lt;sup&gt;-1&lt;/sup&gt;</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Coefficient Estimate</td>
<td>DF</td>
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<tr>
<td>Intercept</td>
<td>0.2253</td>
<td>1</td>
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<tr>
<td>X&lt;sub&gt;1&lt;/sub&gt;</td>
<td>-0.0175</td>
<td>1</td>
</tr>
<tr>
<td>X&lt;sub&gt;2&lt;/sub&gt;</td>
<td>0.1094</td>
<td>1</td>
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<td>X&lt;sub&gt;3&lt;/sub&gt;</td>
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<td>1</td>
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<td>-0.0140</td>
<td>1</td>
</tr>
<tr>
<td>X&lt;sub&gt;1&lt;/sub&gt;X&lt;sub&gt;3&lt;/sub&gt;</td>
<td>-0.0080</td>
<td>1</td>
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<tr>
<td>X&lt;sub&gt;2&lt;/sub&gt;X&lt;sub&gt;3&lt;/sub&gt;</td>
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<td>1</td>
</tr>
<tr>
<td>X&lt;sub&gt;1&lt;/sub&gt;X&lt;sub&gt;1&lt;/sub&gt;X&lt;sub&gt;2&lt;/sub&gt;</td>
<td>-0.0248</td>
<td>1</td>
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<tr>
<td>X&lt;sub&gt;1&lt;/sub&gt;X&lt;sub&gt;2&lt;/sub&gt;X&lt;sub&gt;3&lt;/sub&gt;</td>
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<td>1</td>
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<td>X&lt;sub&gt;1&lt;/sub&gt;X&lt;sub&gt;3&lt;/sub&gt;X&lt;sub&gt;2&lt;/sub&gt;</td>
<td>0.0445</td>
<td>1</td>
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**Table 6**: Results of RSM optimization

<table>
<thead>
<tr>
<th>Inoculum concentration (g L&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>Light intensity (µmol m&lt;sup&gt;-2&lt;/sup&gt; s&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>S/V ratio (m&lt;sup&gt;-1&lt;/sup&gt;)</th>
<th>Volumetric productivity, g L&lt;sup&gt;-1&lt;/sup&gt; day&lt;sup&gt;-1&lt;/sup&gt;</th>
<th>Growth rate, day&lt;sup&gt;-1&lt;/sup&gt;</th>
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<tbody>
<tr>
<td>Pred</td>
<td>Exp</td>
<td>Pred</td>
<td>Exp</td>
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</tr>
<tr>
<td>0.351</td>
<td>180</td>
<td>112</td>
<td>0.529</td>
<td>0.542</td>
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<tr>
<td>0.739</td>
<td>0.785</td>
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</table>

**Table 7**: Comparison of biomass productivity and growth rate of *Arthrospira platensis* obtained from this study with others research’s
<table>
<thead>
<tr>
<th>Algae type</th>
<th>Type of photobioreactor</th>
<th>S/V ratio m⁻¹</th>
<th>Volumetric productivity, $P_V$ (g L⁻¹ day⁻¹)</th>
<th>Growth rate, $\mu_{\text{max}}$ (day⁻¹)</th>
<th>References</th>
</tr>
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<tbody>
<tr>
<td><em>A. platensis</em></td>
<td>HoP</td>
<td>194</td>
<td>0.86</td>
<td>0.988</td>
<td>da Silva <em>et al.</em> 2016</td>
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<td></td>
<td>HeP</td>
<td>388</td>
<td>1.12</td>
<td>0.750</td>
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<tr>
<td><em>M. aeruginosa</em></td>
<td>ABCPBR</td>
<td>90</td>
<td>0.163</td>
<td>-</td>
<td>El-Mekkaoui <em>et al.</em> 2020</td>
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<tr>
<td><em>Chlorella pyrenoidosa</em></td>
<td>- MTA PBR</td>
<td>78</td>
<td>0.107</td>
<td>-</td>
<td>Sukacová <em>et al.</em> 2021</td>
</tr>
<tr>
<td></td>
<td>- H-T PBR</td>
<td>125</td>
<td>0.147</td>
<td>-</td>
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<tr>
<td><em>A. platensis</em></td>
<td>CAPBR</td>
<td>112</td>
<td>0.542</td>
<td>0.785</td>
<td>Present study</td>
</tr>
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</table>

CAPBR: Conical annular Photobioreactor; HoP: Horizontal Photobioreactor; HeP: Helicoidal Photobioreactor; ABC PBR: airlift bubble column photobioreactor; MTA PBR: MultiTubular Airlift photobioreactor; H-T PBR: Helical-Tubular photobioreactor

![Fig. 1: Schematic of conical annular photobioreactor (CAPBR)](image)

![Fig. 2: Plot of predicted values versus to actual values for (a) volumetric productivity and (b) growth rate](image)
Fig. 3: Normal plot of residual for (a) volumetric Productivity and (b) growth rate

Fig. 4: 3D surface plots of the volumetric productivity at (a) light intensity and inoculum concentration; (b) light intensity and S/V ratio; (c) inoculum concentration and S/V ratio
Fig. 5: 3D surface plots of the Growth rate at (a) light intensity and inoculum concentration; (b) light intensity and S/V ratio; (c) inoculum concentration and S/V ratio

Fig. 6: Effect of light intensity and S/V ratio on biomass concentration
Enhancing biomass production by the optimization of process parameters of microalgae (Arthrospira platensis) in conical annular photobioreactor using response surface methodology

Declarations of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Reference


Danesi, E.D.G.; Rangel-Yagui, C.O.; Sato, S. de Carvalho, J.C.M. (2011). Growth and content of Spirulina platensis biomass chlorophyll cultivated at different values of light intensity and temperature using different nitrogen sources, Brazilian Journal of Microbiology.,

Fig. 7 : Effect of light intensity and S/V ratio on chlorophyll content


Enhancing biomass production by the optimization of process parameters of micro algae (Arthrospira platensis) in conical annular photobioreactor using response surface methodology


