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Harnessing of whey and CO_2 for the production of *Arthrospira* (*Spirulina*) *platensis* microalgae biomass: a circular economy approach

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ABSTRACT. The production of microalgae biomass has a high cost due to the composition of the culture medium; therefore, the cultivation of *Arthrospira* (*Spirulina*) *platensis* using carbon dioxide (CO_2) and whey could be considered an effective method of obtaining microalgae biomass while reducing greenhouse gas emissions and a sustainable solution for the treatment of dairy industry wastewater. As a result, the microalgae biomass produced has the potential to become a valuable source of energy (biodiesel), food (for humans or animals), and other compounds such as vitamins and pigments. According to circular economy and development practices, this paper aimed to investigate the growth of *Arthrospira platensis* biomass using CO_2 and whey as a substitute for culture medium. The effects of different whey dilution and culture conditions on biomass productivity, nutrients, and chemical oxygen demand (COD) removal were examined in batch experiments. The results show that the highest biomass production (3.31 $g{\cdot}L^{-1}$) was achieved in 19-day experiments using a modified Schlösser culture medium with 500 mL CO_2 as a carbon source. In the cultures with whey, removal of organic load percentages of 89.16%, 98.88%, and 97.76% was reached on the tenth day in the cultures with 1, 5, and 9 g $COD{\cdot}L^{-1}$, respectively. The highest lipid content of 7.07 g lipids 100 g^{-1} biomass was reached using 500 mL of CO_2 as a carbon source, demonstrating that subjecting the culture to a nutrient deficit causes an accumulation of lipids as long as there is light and CO_2 available.

Keywords: *Arthrospira* (*Spirulina*) *platensis*; carbon dioxide (CO_2); whey; environmental pollution; biomass production; lipid production.

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Introduction

Humans face serious environmental problems, such as climate change and pollution, resulting in the deterioration and depletion of natural resources. Human activity is the main cause of the phenomenon of environmental pollution. Water is one of the most important natural resources on the planet. It is a vital and indispensable liquid for human life and feedstock in numerous industries like pharmaceuticals, electronics, petrochemicals, agrochemicals, food, and the domestic sector (Abdelfattah et al., 2023). From these sectors, every year, it is estimated that there is a global generation of around 330 $x10^9$ m^3 to 380$x10^9$ m^3 of wastewater (Amaro, Salgado, Nunes, Pires, & Esteves, 2023; Díaz et al., 2022; Jones, Van Vliet, Qadir, & Bierkens, 2021; Zhang et al., 2023), with high concentrations of contaminants such as carbon (organic and inorganic), phosphorus, and nitrogen. An increase of 20 to 30% in the world water consumption is expected for the year 2050, leading to wastewater discharges (Sisman-Aydin, 2022). Of this annual wastewater production, only 52% is treated, and the rest (around 170$x10^9$ m^3) is discharged into natural bodies of water (Amaro et al., 2023).

Dairy factory is a vital part of the food sector. Nevertheless, this industry generates much wastewater, i.e., approximately 90% of the milk used is eliminated as wastewater in the form of whey. Whey is the liquid resulting from the precipitation and removal of casein during the cheese-making process. Around 11 million tons of whey are produced yearly worldwide (Athanasiadou, Klontza, Dimitriou-Christidis, Fountoulakis, & Lekkas, 2023), and it is estimated that Global milk production for the year 2030 to grow to1060 million tons. Considering that 30% of the world's milk is used for cheese production. Thus, it is possible to calculate the

whey generated from the cheese-making process, will be around 3180 million tons in 2031. (Ozcelik et al., 2024). This represents a serious environmental pollution issue. Since a thousand liters of whey have the polluting power to the wastewater produced in one day by 450. The polluting power of whey arises due to its high organic load. Whey retains about 55% of the total nutrients in milk, among which lactose, soluble proteins, fats, salts, and vitamins such as B_{12} stand out (Montalvo-Salinas, Ruiiz-Terán, Luna-Solano, & Cantú-Lozano, 2018). This large nutrient content generates approximately 30 to 50 g of biochemical oxygen demand (BOD) and 60 to 80 g chemical oxygen demand (COD) for each liter of whey (Montalvo-Salinas & Cantú-Lozano, 2018). These values of BOD and COD give an idea of the negative effect of the direct discharge of wastewater from the dairy industry into the environment without prior treatment. Nutrient-rich whey can be used or removed in several ways, for instance by means of a membrane filtration process. In this process, the retained whey proteins are often marketed as protein powders, protein bars, and whey drinks for sports nutrition. On the other hand, whey permeates (the liquid remaining after filtration), which is rich in lactose, can be valorized by converting it into lactose powder or higher-value products such as organic acids, alcohols, polymers, monosaccharides or galactooligosaccharides. Although these technologies have proven effective, they are considered expensive and energy-intensive due to their dependence on enzymes or microorganisms (Ozcelik et al., 2024).

Another serious issue is the constant increase in CO_2 concentration levels. Carbon dioxide (CO_2) is the most important gas contributing to the greenhouse effect and represents about 76% of the total greenhouse gases. Atmospheric CO_2 concentration increased from 280 ppm in the pre-industrial period (1850-1900) to 403 ppm in January 2016, reaching 415 ppm by November 2021 (Cui et al., 2023; Miranda, Sáez, Hoyos, Gómez, & Vargas, 2021; Montalvo et al., 2023). The CO_2 concentration reached 418.51 ppm in September 2023 (McGee, 2023). The increase in CO_2 emissions is mainly due to human activities such as burning fossil fuels, the transportation sector, energy generation, deforestation, and industrial processes. These emissions are recognized as the main cause of greenhouse gases, which cause climate change and global warming (Bergougui, 2024). Given the above, reducing CO_2 emissions has become one of the main goals for highly industrialized nations and private industries worldwide. This goal establishes an international framework to avoid severe climate change, keeping global warming below 2 °C (Serafin, Dziejarski, & Sreńscek-Nazzal, 2023).

In this context, proposing a process that uses microalgae cultivation offers several advantages over traditional technologies due to the rapid absorption of nutrients in whey, the reduction of CO_2 emissions given its autotrophic metabolism, and possible cost savings.

Microalgae can be defined as a set of microorganisms with photosynthetic capacity. In general, they are unicellular microorganisms such as cyanobacteria in the form of filaments. These microorganisms are essential to regulating ecosystems due to their exceptional capacity for CO_2 fixation and wastewater treatment (Yu et al., 2023). From microalgae, we can obtain compounds of interest, such as biomass, pigments, enzymes, sugars, lipids, sterols, antioxidants, carotenoids, and vitamins (Goshtasbi et al., 2023; Uma et al., 2023). These compounds are useful in several areas, including pharmaceuticals, cosmetics, chemicals, fish farming food, agriculture industries, and biofuel (Alishah Aratboni et al., 2023; Ibrahim et al., 2023). These microorganisms are capable of growing in marine and freshwater environments. Within the microalgae group is *Spirulina* (trade name), which includes two main species of cyanobacteria: *Arthrospira platensis* and *Arthrospira maxima* (Lafarga, Fernández-Sevilla, González-López, Acién-Fernández, 2020), being the most cultivated in the world, its production can be carried out cyclically (cultivation-harvest-obtaining bioproducts). In addition, using some types of waste to cultivate *Spirulina* reduces production costs and the environmental impact (Costa et al., 2019).

Arthrospira (*Spirulina*) *platensis* is an autotrophic cyanobacterium known as blue-green algae. Its name is given by the nature of its helical or spiral filaments. This microalgae is found in tropical regions and grows naturally in shallow water bodies in the presence of sodium bicarbonate and alkaline media (pH 10-11) with high salinity (NaCl) (Ilieva, Zaharieva, Najdenski, & Kroumov, 2024). The microalgae *Spirulina platensis* is certified as Generally Recognized As Safe (GRAS); therefore, the biomass obtained from these cyanobacteria is used in the preparation of food and medicines for humans and animals (Fernandes et al., 2023). The great interest in the biomass production of the microalgae *Spirulina platensis* is due to its appreciable high content of protein (~70%) with high digestibility and the balanced content of essential amino acids, fatty acids, and active substances such as vitamins, minerals, and phenols (Al-Deriny et al., 2020; AlMulhim, Virk, Abdelwarith, & AlKhulaifi, 2023). According to the FAO statistical data mentioned by Hamidi, Mohammadi, Mashhadi, and Mahmoudnia (2023), in 2010, the global microalgae biomass production was 93,756 tons; later,

in 2018, the global microalgae biomass production was 87,000; and by 2019, it reached 56,465 tons, which were produced by the following countries: China (97.16%), Chile (1.6%), France (0.37%), Greece (0.25%), Tunisia (0.25%), Burkina Faso (0.25%), Central African Republic (0.09%), Chad (0.04%), Bulgaria (0.005%), and Spain (0.003%). Of this production, the cyanobacterium *Spirulina platensis* represents 96.56% of the world's production of microalgae biomass, while the four species of green microalgae represent the remainder (0.44%): *Haematococcus pluvialis* (242 tons, 0.429%), *Chlorella vulgaris* (4.77 tons, 0.008%), *Tetraselmis* sp. (1.45 tons, 0.003%), and *Dunaliella salina* (0.22 tons, 0.00004%).

Pereira et al. (2019) explored the cultivation of microalgae *Spirulina platensis* using whey. They demonstrated that microalgae *Spirulina platensis* can grow in mixotrophic cultures. These authors used Zarrouk's synthetic medium and nutrient-rich whey as culture medium, reaching a maximum biomass production of 2.98 $g{\cdot}L^{-1}$ when using 5% whey. The results showed that organic and inorganic carbon sources increase biomass and carbohydrate productivity in *Spirulina platensis* when whey is added to the culture medium.

Using mathematical models to predict microalgae growth enables the anticipation of cell microalgae development in various environments and under different conditions. In addition, it allows for more effective planning and control in both industrial and laboratory processes. In this study, two widely used unstructured models, the Logistic and Modified Gompertz models, were evaluated to describe the growth of microalgae *Spirulina platensis*. The Logistic model describes microbial population growth as a function of growth rate, initial biomass, maximum biomass, and time, assuming sufficient substrate. The Gompertz model was originally used to describe the distribution of ages in human populations and was later modified (20th century) to model microbial growth (Wang & Guo, 2024). The Modified Gompertz model describes growth as a function of maximum biomass concentration, productivity, and time. It is ideal for microorganisms with extended lag phases and slower growth rates, as it assumes an exponential decrease in growth rate over time (Guzmán-Armenteros, Villacís-Chiriboga, Guerra, & Ruales, 2024).

Finally, circular bioeconomy is defined as creating various renewable biological resources and converting them into high-value products such as food, feed, biochemicals, and bioenergy (Ahmed et al., 2023; Fal, Smouni, & Arroussi, 2023). Therefore, this study focuses on integrating agro-industrial waste, such as whey and carbon dioxide, into sustainable resources to obtain value-added products such as algal biomass.

Material and methods

Figure 1 shows the general methodology of this study, which includes the preparation of the microorganism culture media, the experimental conditions applied, the analyses performed, and the growth kinetic parameters. In later sections, each stage of the experiment is detailed.

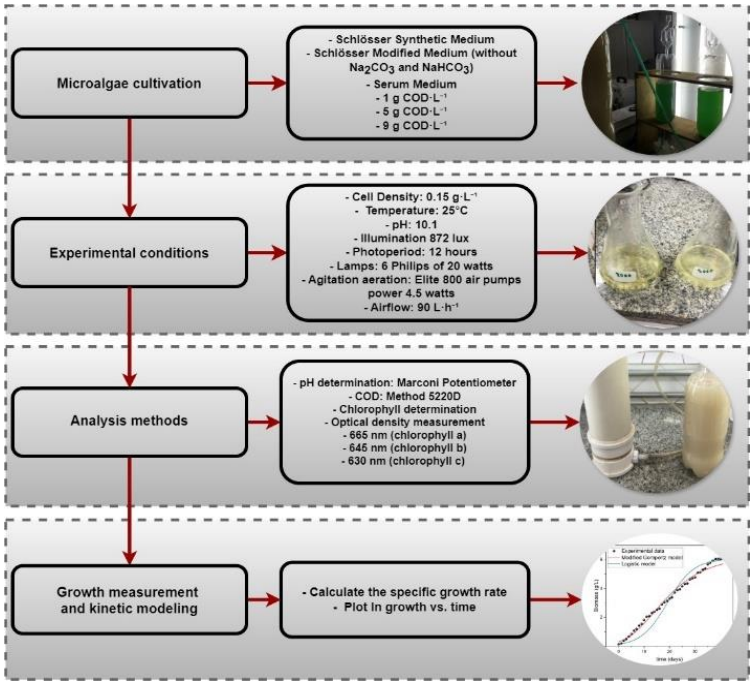

Figure 1. Schematic diagram of experimental development for the cultivation of microalgae *Spirulina platensis*.

Microorganisms and culture media

For this research, the cyanobacterium *Spirulina platensis* strain was cultivated in different mediums: a synthetic Schlösser medium (Schlösser, 1982), a modified Schlösser medium in which carbon-rich compounds (Na_2CO_3, $NaHCO_3$) were not added to induce the microalgae to fix carbon from CO_2; and in a medium composed of whey with different concentrations of chemical oxygen demand (1, 5, and 9 g COD·L^{-1}) in an effective volume of 940 mL. The different COD concentrations of whey were obtained using Equation 1:

$$C_1V_1 = C_2V_2 \quad (1)$$

where: C_1 is the initial concentration, V_1 is the initial volume of the solution before dilution, C_2 is the final concentration of the solution after dilution, and V_2 is the final volume of the solution after dilution.

Table 1 shows the Schlösser medium composition. This medium was utilized due to its rich mineral composition.

Table 1. Composition of Schlösser medium.

Substance	Concentration (g·L^{-1})
$NaHCO_3$	13.8
Na_2CO_3	4.03
K_2HPO_4	0.50
$NaNO_3$	2.50
K_2SO_4	1.00
NaCl	1.00
$MgSO_4·7H_2O$	0.20
$CaCl_2·2H_2O$	0.04
*PIV solutions metals	6 mL
**Chu micronutrient solution	1 mL
*PIV solutions metals	
Na_2EDTA	0.750
$FeCl_3·6H_2O$	0.097
$MnCl_2·4H_2O$	0.041
$ZnCl_2$	0.005
$Na_2MoO_4·2H_2O$	0.004
$CoCl_2·6H_2O$	0.002
**Chu micronutrient solution	
Na_2EDTA	0.050
H_3BO_3	0.618
$CuSO_4·5H_2O$	0.020
$ZnSO_4·7H_2O$	0.044
$CoCl_2·6H_2O$	0.020
$MnCl_2·4H_2O$	0.013
$Na_2MoO_4·2H_2O$	0.013

Experimental conditions

Experimental runs were carried out at a cell density of 0.15 g·L^{-1}, a constant temperature of 25 °C, a pH of 10.1, and lighting of 872 lux under photoperiods of 12 hours. Six Philips lamps of 20 watts provided the lighting. Cultures were agitated by aeration using Elite 800 model air pumps with a power of 4.5 watts and an airflow of 90 L·h^{-1}.

Analysis methods

The pH was determined with a Marconi potentiometer. The organic load was determined by the method 5220D Chemical Oxygen Demand/Standard Methods (Standard Methods Committee of the American Public Health Association, 2017). The percent of organic matter removal was calculated from COD measurements as follows (Atiyah, Muallah, & Abbar, 2024):

$$\text{COD removal } (\%) = \frac{COD_i - COD_t}{COD_i} \cdot 100 \quad (2)$$

where: COD_i corresponds to the initial value, and COD_t is the value at time t. The biomass was determined by the dry weight method. The determination of chlorophyll was carried out using the following methodology: 10 mL of the suspension of microalgae *Spirulina platensis* were taken and centrifuged at 3,200 rpm for 25 min.

at room temperature. Subsequently, the supernatant was discarded, and 5 mL of methyl alcohol was added to the sedimented biomass, stirring vigorously to suspend the biomass. The mixture of biomass suspension and methyl alcohol was covered with aluminum foil to prevent photooxidation of the chlorophyll. This mixture was stored at 5 °C for 24 hours. After this time, centrifugation is performed again at 3,200 rpm for 25 min. Once the pigments were extracted with the second centrifugation, the optical density was determined in a spectrophotometer at three wavelengths: 665 nm for chlorophyll a, 645 nm for chlorophyll b, and 630 nm for chlorophyll c. The quantification of chlorophyll concentration was expressed in $mg{\cdot}L^{-1}$ of methanol and the correlations of Parsons and Strickland (Parsons' & Strickland, 1963) were used.

$$\text{Chlorophyll a} = (11.6)(D665) - (0.14)(D630) - (1.31)(D645) \quad (3)$$

$$\text{Chlorophyll b} = (20.7)(D645) - (4.34)(D665) - (4.42)(D630) \quad (4)$$

$$\text{Chlorophyll c} = (55.0)(D630) - (16.3)(D645) - (4.65)(D665) \quad (5)$$

Total lipids were determined using the methodology proposed by Bligh and Dyer (1959). Lipid content was expressed by Equation 6:

$$\frac{\text{glipids}}{100\text{gbiomass}} = \frac{\text{w}_\text{f} - \text{w}_\text{i}}{\text{biomass}} \quad (6)$$

where: w_f is the final weight of the tube with lipids, w_i is the initial weight of the tube without lipids, and biomass is the dry weight of the biomass used.

Growth measurement and kinetic modeling

The value of the specific growth rate can be calculated from the slope of the graph, which plots the natural logarithm of *Spirulina platensis* growth against time. It is also calculated using the mathematical expression below:

$$\mu = \frac{\ln X_i - \ln X_0}{t_i - t_0} \quad (7)$$

where: X_i represents the mass of biomass on the ith day (t_i), and X_0 is the mass of biomass on the initial day (t_0). The double time (T_d), which indicates the generation time, was calculated using Equation 8:

$$T_d = \frac{0.693}{\mu} \quad (8)$$

The value of n is the number of generations, which was obtained by dividing the period of logarithmic growing time by the generation time.

$$n = \frac{\Delta t}{T_d} \quad (9)$$

Productivity (φ) is evaluated as the biomass production per unit time per unit culture volume:

$$\varphi = \frac{W_b - W_{b0}}{t_i - t_0} \quad (10)$$

where: W_b is the weight of biomass on the ith day (t_i), and W_{b0} is the weight mass of biomass on the initial day (t_0). Finally, sigmoid growth models were fitted to the experimental data. The adjusted models were the modified Gompertz (Equation 11) and logistic (Equation 12) models:

$$X = X_0 + \left[X_{max} \cdot e\left[-e\left(\frac{r_m \cdot e}{X_{max}}\right)(t_l - t) + 1\right]\right] \quad (11)$$

$$X = \frac{X_0 \cdot e(\mu_{max} t)}{1 - \left[\left(\frac{X_0}{X_{max}}\right)(1 - e(\mu_{max} t))\right]} \quad (12)$$

where: X is the cell concentration, X_0 is the initial cell concentration, X_{max} is the maximum cell concentration, μ_{max} is the maximum growth rate, r_m is the maximum cell production rate, and t_l is the lag phase. As for the COD degradation, Weibull distribution was used (Equation 13). The Weibull model was initially developed to describe the failure of a system under stress conditions over time. However, it has been recently used to model chemical degradation kinetics (Wang et al., 2021).

$$COD_t = COD_i e\left[-\left(\frac{t}{\alpha}\right)^{\beta}\right] \quad (13)$$

where: COD_t is the chemical oxygen demand at time t, COD_i is the initial chemical oxygen demand, α is the scale parameter corresponding to the time required for a significan reduction, β is the shape parameter that indicates whether the curve is concave (β<1), convex (β>1) or straight line (β=1), and t is time.

Results and discussion

The microalgae cells were observed in the optical microscope with a 40x objective. Figure 2 shows the filamentous morphology of microalgae *Spirulina platensis*. It is possible to observe that the microalga has a filamentous structure, made up of multicellular cylindrical trichomes spiral-shaped, which are arranged in the form of an open helix.

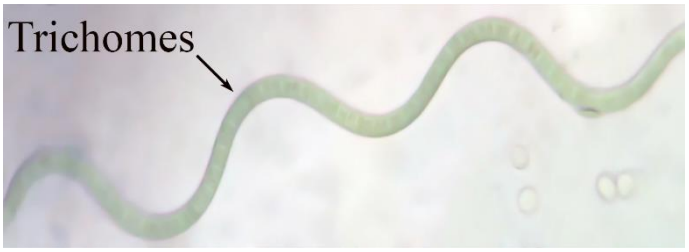

Figure 2. Microscopic morphology of *Spirulina platensis* (40x).

Initially, the strategy was to inoculate the microalgae *Spirulina platensis* in the Schlösser synthetic medium, shaking only with air, to observe the development of microalgae in a medium suitable for its growth. Figure 3 shows the representative growth curve, in which it can be seen that the cyanobacteria adapted favorably to the environment, reaching 6.33 g·L^{-1} of biomass, a value that is higher compared to data already reported by Arahou, Hassikou, Arahou, Rhazi, and Wahby (2021), Saxena et al. (2022), and Gomaa, Ali, and Hifney (2023). As can be seen, growth continued until day 38, then the stationary phase began. This behavior can be explained by the effective agitation provided in the culture, which prevented the sedimentation of nutrients and cyanobacteria. It is important to prevent sedimentation for the following reasons: I) To ensure adequate nutrient mixing. If microalgae settle, they may remain in areas with insufficient nutrient concentrations. II) To avoid the formation of anoxic zones with little or no oxygen, which can be detrimental to microalgae growth. III) To maintain a homogeneous culture, preventing areas of high and low cell density that may affect the system's overall productivity.

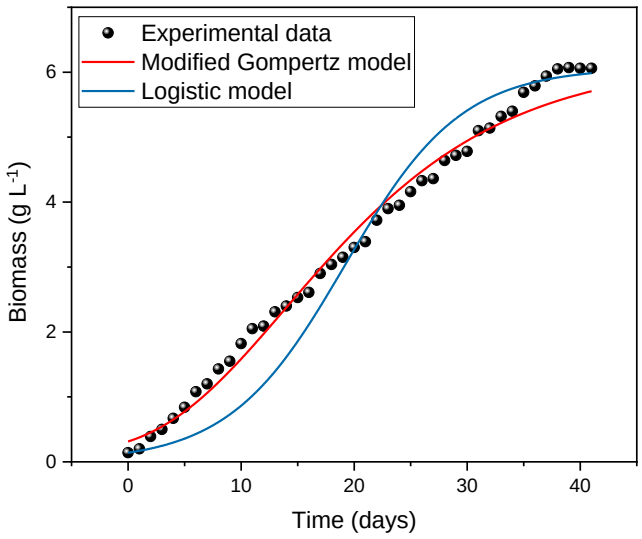

Figure 3. Growth curve of microalgae *Spirulina platensis* in Schlösser medium.

The chlorophyll present in the biomass of *Spirulina platensis* grown using the synthetic Schlösser medium was quantified, and it was determined that the biomass obtained during this experiment consisted of 9.4526 mg·L^{-1} of chlorophyll a, 26.7001 mg·L^{-1} of chlorophyll b, and 70.7750 mg·L^{-1} of chlorophyll c. Therefore, the total value was equivalent to 106.9277 mg·L^{-1} of chlorophyll present in this microalga.

Figure 4 shows the growth of microalgae *Spirulina platensis* with the modified Schlösser medium (Na_2CO_3 and $NaHCO_3$ were not added) to induce carbon fixation from CO_2. As can be seen, the microalgae have good cellular development with a minimal adaptation phase when CO_2 is used as a carbon source (500 mL·h^{-1}), reaching a biomass of 2.71 g·L^{-1}. The constant addition of CO_2 results in the pH decreasing to values around 7.5-8 in the first 24 hours of cultivation due to the dissolution of the gas in the medium, and subsequently, the pH remaining constant during the following days of cultivation. For the modified Schlösser culture, without adding CO_2 as a carbon source, it is impossible to identify a defined growth phase reaching the stationary phase after 20 days. The average biomass obtained in these cultures was around 0.9 g·L^{-1}. In these cultures (without CO_2 contribution), the pH increased to 11-11.8. According to Vives, Ugás, and Yero (2021), as CO_2 from the medium is consumed by algae, the pH increases due to equilibrium considerations. In contrast, when there is organic substrate to be consumed, the pH decreases due to the production of CO_2. Pedraza (1989) mentioned that as the microalgae consume the CO_2 dissolved in the medium, they modify the levels of carbonic acid, increasing pH.

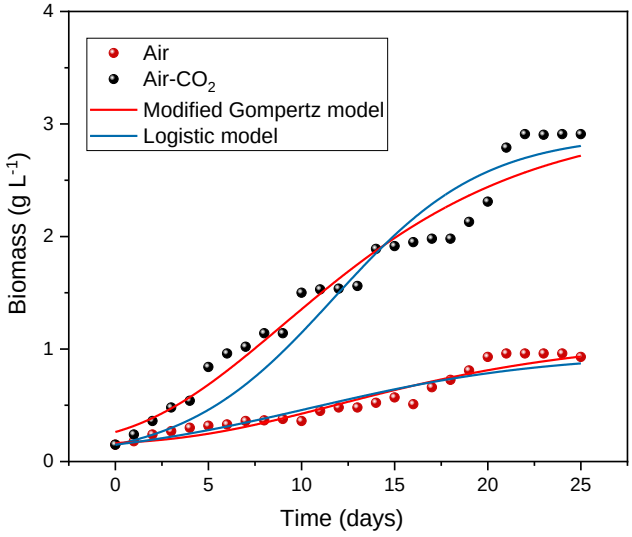

Figure 4. *Spirulina platensis* grown in modified Schlösser medium with and without the addition of CO_2.

The advantage of CO_2 injection into the culture medium is that it helps establish better conditions of photosynthesis and metabolism within microalgae cells for reasons: i) increased carbon availability enhances the photosynthetic activity because the CO_2 acts as a carbon source in the photosynthetic process; ii) dissolved CO_2 forms carbonic acid, dissociating into bicarbonate and protons. It helps maintain an adequate pH, improving mineral solubility and affecting cellular metabolism and biomass composition; and iii) under low CO_2 concentration conditions, microalgae undergo photorespiration, which consumes energy and carbon without producing carbohydrates. As CO_2 levels increase, photorespiration is reduced, and light use efficiency is improved. Therefore, the growth of the microalgae was evaluated using the modified Schlösser medium, and different CO_2 flows to determine the CO_2 concentration at which the microalgae present the highest biomass production. Figure 5 shows the growth of the cyanobacterium *Spirulina platensis* at three different CO_2 feed concentrations. CO_2 feeding was carried out every 72 hours for an average period of around 30 minutes. In the graph, it can be seen that there is no significant difference between the cultures fed with 250 mL and 500 mL of CO_2. However, the highest microalgae production biomass was obtained in the culture fed with 500 mL of CO_2, reaching a maximum production of 3.31 g of biomass·L^{-1} after 19 days, a value that exceeded the biomass obtained at the same time from the culture in standard Schlösser synthetic medium. The harvested biomass of *Spirulina platensis* with the injection of 500 mL of CO_2 was higher than the biomass obtained by Kim et al. (2013) when injecting 1% of CO_2 into the culture medium. In comparison with other studies where the purpose is to use microalgae for CO_2 fixation, Morais and Costa, (2007), working with *Chlorella kessleri* and *Scenedesmus obliquus*, found that *S. obliquus* presented a maximum growth in the presence of 12% CO_2 (1.14 g·L^{-1}), while *C. kessleri* reached a maximum of 1.45 g·L^{-1}in the presence of 0.038% CO_2. These authors mentioned that the relatively high resistance to CO_2 and high growth rate of *S. obliquus* indicate that these microalgae could utilize the emissions of the coal- or oil-fired thermoelectric power

industry, where the concentration of CO_2 is much higher than that of the atmosphere. On the other hand, Chang and Yang (2003) reported a maximum production of 1.8 g·L^{-1} of biomass using the microalgae *Chlorella sp*. under aeration of 15% CO_2 for 10 days.

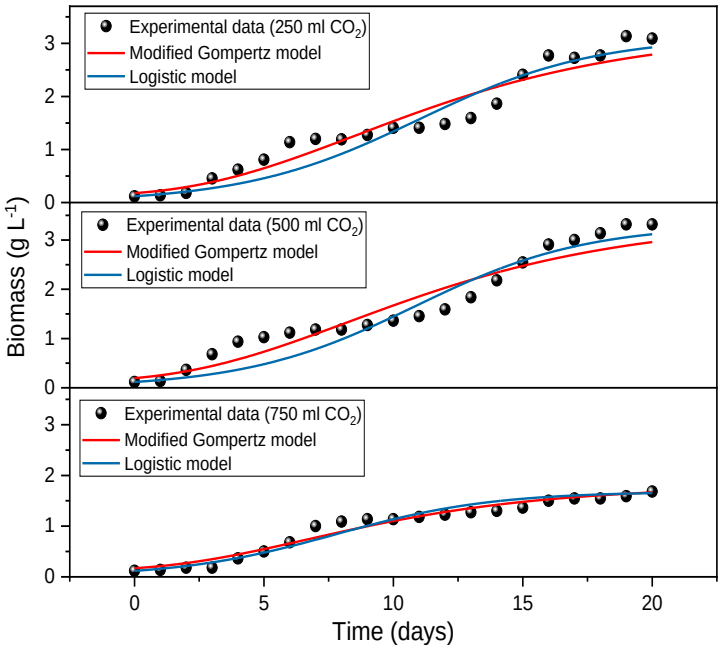

Figure 5. *Spirulina platensis* biomass under different concentrations of CO_2.

Using whey as a growth medium for microalgae cultivation reduces costs and freshwater usage and transforms the high organic load in whey into valuable biomass. Figure 6 shows the growth of biomass at different COD concentrations. The figure shows that the highest biomass production was obtained at an initial concentration of 5 g COD·L^{-1}, reaching a biomass of 1.91 g·L^{-1} on the seventh day, this culture presenting the greatest removal of organic load. The S_0 = 9 g COD·L^{-1} culture presented a longer lag phase, and biomass production was lower than the other culture. This behavior is explained by the fact that there is growth inhibition due to the high concentration of substrate.

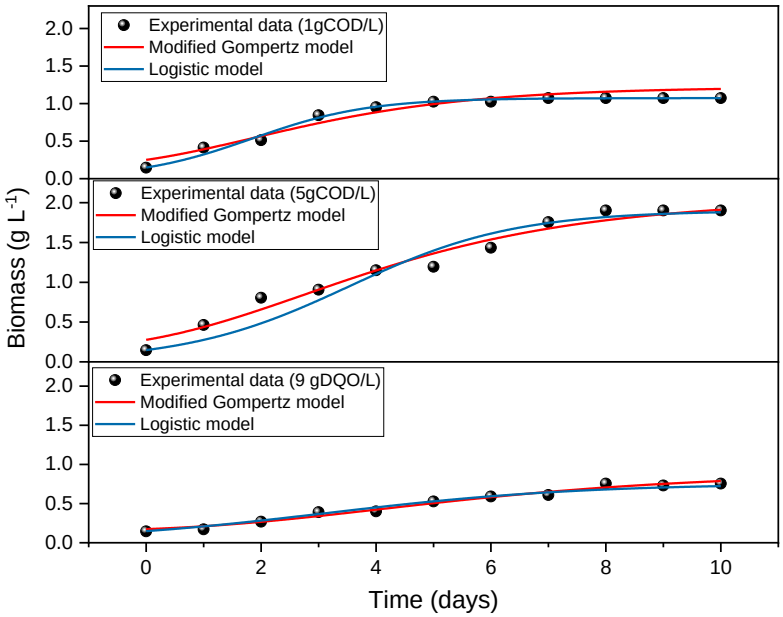

Figure 6. *Spirulina platensis* biomass under different COD concentrations.

Organic load removal percentages of 89.16, 98.88, and 97.76% were reached on the tenth day in the cultures with 1, 5, and 9 g COD·L^{-1}, respectively. Compared to other works, Bucci et al. (2024) achieved 100%

removal of COD present in whey used as a growth medium for microalgae culture. Riaño, Blanco, Becares, and García-González (2016) achieved a removal efficiency of 48% of the total organic load in whey, reaching a microalgal biomass production between 0.61-1.10 g VSS·L^{-1}. The Weibull model was used to describe COD degradation. In Figure 7, it can be seen that the Weibull model adequately fitted the experimental data (R^2 > 0.95). The Weibull models found experimentally for COD removal are presented in Equations 14, 15, and 16 for the media containing 9, 5, and 1 g COD, respectively. The values for β (>1) indicate a slow degradation followed by a rapid COD degradation rate. The times (in days) that achieved significant degradation of organic matter were 2.84, 3.52, and 5.63 for organic loads of 1, 5, and 9 g COD, respectively.

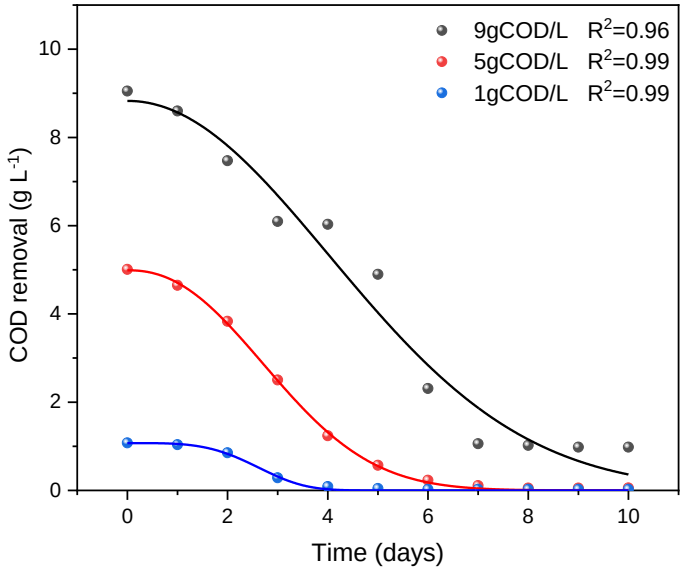

Figure 7. COD degradation modeled by Weibull model.

$$COD_t = 9.05e\left[-\left(\frac{t}{5.63}\right)^{2.02}\right] \qquad R^2 = 0.96 \qquad (14)$$

$$COD_t = 5.01e\left[-\left(\frac{t}{3.52}\right)^{2.25}\right] \qquad R^2 = 0.99 \qquad (15)$$

$$COD_t = 1.07e\left[-\left(\frac{t}{2.84}\right)^{3.83}\right] \qquad R^2 = 0.99 \qquad (16)$$

The key purpose of fitting kinetic models to microalgae growth is to understand how microalgae respond to different environmental conditions and how these conditions affect their growth rate. On the other hand, adjusting different growth kinetic models to experimental data can help find the model that best predicts and describes microalgae growth under the environmental conditions studied. Table 2 presents the kinetic growth parameters of the microalgae *Spirulina platensis* in each medium used and the models adjusted to the experimental data. In the table, it can be seen that the model that best fits the experimental data is the Gompertz model. The Gompertz model fitted the growth kinetic data better compared to the logistic model due to the following reasons: i) the sigmoid curve produced by the Gompertz model is asymmetric, which explains that the exponential growth phase and the deceleration phase are non-symmetric, *i.e.*, the initial growth of the culture is slow, followed by a growth acceleration and finally a deceleration before reaching the stationary phase (see Figure 3); ii) the model features the lag phase parameter (t_l), which allows for a better representation of the adaptation period of microalgae before they begin to grow exponentially (see Equation 11).

The non-addition of carbon-rich compounds (Na_2CO_3, $NaHCO_3$) in the modified Schlösser medium decreases the growth rate. On the other hand, adding CO_2 to the modified Schlösser medium increases the growth rate, with a flow of 500 mL of CO_2 being the optimal concentration. Using whey as a growth medium significantly increases the growth rate, reaching a high productivity of 0.21 g·L^{-1} day^{-1} on the eighth day. However, the rapid COD removal in the whey does not allow for achieving high dry biomass production.

Table 2. Growth kinetic parameters of *Spirulina platensis* biomass at different growth mediums.

Medium	Growth kinetic parameters				Models	R^2
	μ (d^{-1})	T_d (d)	n	φ (g L^{-1}·d)		
Schlösser	0.099	6.99	5.43	0.15	Gompertz $X = 0.14 + \left[6.05 \cdot e\left[-e\left(\frac{0.203 \cdot e}{6.05}\right)(2.96 - t) + 1\right]\right]$	0.98
					Logistic $X = \frac{0.14 \cdot e(0.195t)}{1 - \left[\left(\frac{0.14}{6.05}\right)(1 - e(0.195t))\right]}$	0.92
MS/a	0.088	7.87	2.66	0.04	Gompertz $X = 0.15 + \left[0.96 \cdot e\left[-e\left(\frac{0.042 \cdot e}{0.96}\right)(3.58 - t) + 1\right]\right]$	0.93
					Logistic $X = \frac{0.15 \cdot e(0.159t)}{1 - \left[\left(\frac{0.15}{0.96}\right)(1 - e(0.159t))\right]}$	0.90
MS/CO_2	0.134	5.17	4.25	0.12	Gompertz $X = 0.15 + \left[2.91 \cdot e\left[-e\left(\frac{0.139 \cdot e}{2.91}\right)(1.38 - t) + 1\right]\right]$	0.95
					Logistic $X = \frac{0.15 \cdot e(0.247t)}{1 - \left[\left(\frac{0.15}{2.91}\right)(1 - e(0.247t))\right]}$	0.92
MS/250 mLCO_2	0.172	4.02	4.71	0.15	Gompertz $X = 0.12 + \left[3.13 \cdot e\left[-e\left(\frac{0.185 \cdot e}{3.13}\right)(2.37 - t) + 1\right]\right]$	0.93
					Logistic $X = \frac{0.12 \cdot e(0.295t)}{1 - \left[\left(\frac{0.12}{3.13}\right)(1 - e(0.295t))\right]}$	0.92
MS/500 mL CO_2	0.166	4.17	4.79	0.16	Gompertz $X = 0.12 + \left[3.31 \cdot e\left[-e\left(\frac{0.194 \cdot e}{3.31}\right)(2.00 - t) + 1\right]\right]$	0.92
					Logistic $X = \frac{0.12 \cdot e(0.304t)}{1 - \left[\left(\frac{0.12}{3.31}\right)(1 - e(0.304t))\right]}$	0.91
MS/750 mL CO_2	0.132	5.25	3.80	0.08	Gompertz $X = 0.12 + \left[1.68 \cdot e\left[-e\left(\frac{0.115 \cdot e}{1.68}\right)(1.31 - t) + 1\right]\right]$	0.96
					Logistic $X = \frac{0.12 \cdot e(0.331t)}{1 - \left[\left(\frac{0.12}{1.68}\right)(1 - e(0.331t))\right]}$	0.94
1g COD	0.248	2.79	2.87	0.11	Gompertz $X = 0.14 + \left[1.07 \cdot e\left[-e\left(\frac{0.18 \cdot e}{1.07}\right)(-0.32 - t) + 1\right]\right]$	0.92
					Logistic $X = \frac{0.14 \cdot e(0.98t)}{1 - \left[\left(\frac{0.14}{1.07}\right)(1 - e(0.98t))\right]}$	0.98
5g COD	0.320	2.16	3.69	0.21	Gompertz $X = 0.14 + \left[1.90 \cdot e\left[-e\left(\frac{0.20 \cdot e}{1.90}\right)(-0.02 - t) + 1\right]\right]$	0.97
					Logistic $X = \frac{0.14 \cdot e(0.69t)}{1 - \left[\left(\frac{0.14}{1.90}\right)(1 - e(0.69t))\right]}$	0.93
9g COD	0.205	3.38	2.36	0.07	Gompertz $X = 0.14 + \left[0.75 \cdot e\left[-e\left(\frac{0.08 \cdot e}{0.75}\right)(0.71 - t) + 1\right]\right]$	0.97
					Logistic $X = \frac{0.14 \cdot e(0.451t)}{1 - \left[\left(\frac{0.14}{0.75}\right)(1 - e(0.451t))\right]}$	0.97

*d = days, MS/a = Modified Schlösser/air, MS/CO_2= Modified Schlösser/CO_2

Finally, the extraction of lipids was carried out using 100 mg of dry biomass belonging to the cultures with the highest biomass production (Schlösser synthetic, modified Schlösser/500 mL of CO_2-air, and modified

Schlösser/500 mL of CO_2), performing each extraction in duplicate. Figure 8 shows the results in grams of lipids per 100 g of biomass. It is possible to see in the figure that the culture that is fed only with CO_2 as a carbon source presented the highest lipid content of 7.07 g lipids 100 g^{-1} biomass, which demonstrates that subjecting the culture to a nutrient deficit causes an accumulation of lipids as long as there is light and CO_2 available (Magierek & Krzemińska, 2018; Suparmaniam et al., 2023; Yaakob, Mohamed, Al-Gheethi, Aswathnarayana Gokare, & Ambati, 2021).

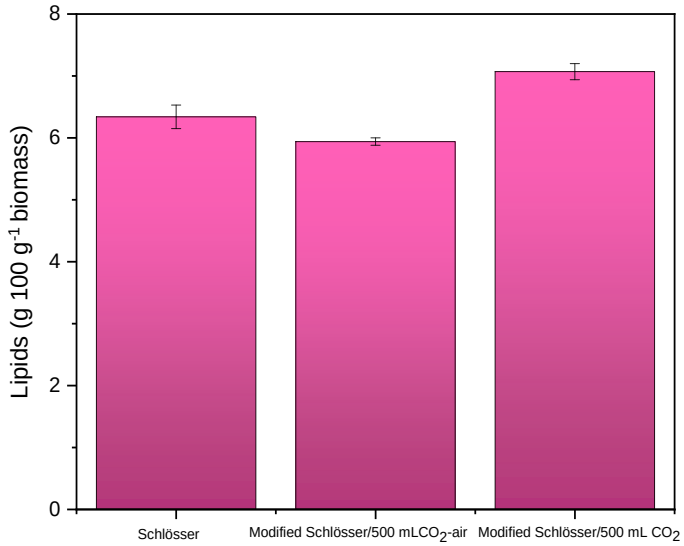

Figure 8. Lipid content in different cultures of microalgae *Spirulina platensis*.

Conclusion

The results suggest that the cultivation of microalgae biomass of *Spirulina platensis* can be considered a promising alternative, environmentally safe, and sustainable technology to capture CO_2 and, at the same time, a strategy for biotreatment of dairy wastewater to reduce the disposal of untreated whey in nature (Garrido-Cardenas, Manzano-Agugliaro, Acien-Fernandez, & Molina-Grima, 2018), as well as the sourcing of biomass that can convert the end products into products with added value. The optimal CO_2 feed was 500 mL, and we found that at concentrations higher than this feed, the growth of microalgae is inhibited by the sudden change in pH. The microalgae *Spirulina platensis* demonstrated a high capacity to reduce the organic load present in whey. The initial concentration of 5 g COD·L^{-1} whey was obtained from the highest biomass production, with a COD removal of 98.88%. The non-addition of carbon-rich compounds (Na_2CO_3, $NaHCO_3$) in the modified Schlösser medium decreases the growth rate of *Spirulina platensis* microalgae. On the other hand, adding CO_2 to the modified Schlösser medium increases the growth rate, with a flow of 500 mL of CO_2 being the optimal concentration. Using whey as a growth medium significantly increases the growth rate, reaching a high productivity of 0.21 g·L^{-1}·day^{-1} on the eighth day. However, the rapid removal of COD in whey does not allow for achieving high dry biomass production. The Gompertz model best fits the microalgae growth kinetic data because it can describe an asymmetric growth curve and represent the lag phase more realistically.

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