SCIENTIFIC EVENTS GATE

The International Innovations Journal of Applied Science

Journal homepage: https://iijas.eventsgate.org/iijas

ISSN: 3009-1853 (Online)

Enhancing Microalgal Biomass and Chemical Composition for Sustainable Wastewater Treatment

Muhja Abdul Kareem Ahmed [1], Hala Ali Turky[2] , Dina Hazim Abddulhameed[3], Muna Mohammed Khayri[4] , Raghda Salim Sultan[5*] , Aya Mohammed Hussein [6]

[1]Department of Biochemistry, Collage of Science, University of Baghdad, Baghdad, Iraq.

[2,3,4,5,6] Department of Biotechnology, Collage of Science, University of Baghdad, Baghdad, Iraq.

ARTICLE INFO

Article history:
Received 21 May. 2024
Revised 2 Jun. 2024,
Accepted 11 Jun. 2024,
Available online 15 Jul. 2024

Keywords:
Biomass production
Chemical Composition
Sustainable solutions
Central Composite Design (CCD)
Wastewater Treatment

ABSTRACT

The research explores enhancing microalgal biomass production and its chemical composition for sustainable wastewater treatment. By using the Central Composite Design (CCD) methodology, nutrient concentrations and microbial components in the growth medium were optimized. Adjustments to light exposure, particularly with red and blue frequencies, significantly improved biomass yield, carbon sequestration, and protein content. Optimal light intensity further enhanced these parameters. A process incorporating on-demand CO_2 supply and controlled pH levels showed potential for increasing biomass concentrations and nutrient utilization efficiency. Results demonstrated a 28% increase in biomass fixation and a 27% increase in biomass production compared to unoptimized conditions. The study emphasizes the effectiveness of integrated approaches in boosting biomass production and optimizing wastewater treatment, offering promising avenues for sustainable bio-based solutions

1. Introduction

In an era marked by escalating environmental concerns, resource scarcity, and a growing global population, the imperative for sustainable solutions is more pressing than ever Biotechnology and biochemistry offer distinct tools and strategies to address these challenges. By integrating biotechnological advancements with a fundamental understanding of biochemical cycles, we can develop sustainable solutions that promote environmental stewardship, economic growth, and social equity (Alkhaddar et al., 2005; Ah-You et al., 2000; Alivisatos, 2004).

However, the high cost of biomass production and low biomass concentration remain significant obstacles to the widespread adoption of microalgae for wastewater treatment.

The aim of this paper to explore how the collaboration between biotechnology and biochemistry can enhance microalgal biomass and chemical composition to make wastewater treatment more efficient and economically viable (Andres et al., 2006).

This study proposes several solutions and innovations to address these challenges:

Nutrient Optimization: By fine-tuning the nutrient inputs necessary for microalgal growth, we aim to improve the economics of biomass production and reduce costs (Baptista

* Corresponding author.
E-mail address: raghda.s@sc.uobaghdad.edu.iq

et al., 2006).

Species-Specific Media: Developing media and environmental conditions tailored to specific microalgal species can enhance growth performance and make the production process more efficient and economically viable (Beaudette et al., 2002).

Advanced Lighting Technologies: Utilizing light-emitting diodes (LEDs) of various wavelengths can provide efficient, long-lasting, reliable, cost-effective, and adjustable light sources for microalgae culture. This approach aims to optimize light conditions to maximize biomass yield and enhance biochemical compositions (Beck-Friis, 2001).

Genetic Enhancements: Efforts to upregulate genes involved in CO_2 fixation, substrate utilization, light availability, and CO_2 mass transfer can significantly increase biomass yield. These genetic enhancements can make the process more sustainable and productive (Beliaeff and Burgeot, 2002).

By focusing on optimizing nutrient inputs, improving light conditions, and enhancing genetic factors, the production of microalgal biomass for sustainable wastewater treatment can be made more efficient and economically viable.

The sustainable livelihoods paradigm is being used by the International Food Policy Research Institute (IFPRI) to investigate how agricultural research affects poverty. This multidisciplinary approach provides insights into the nature of poverty, the impact of capital assets on livelihood strategies, social conflict, market shock susceptibility, and vulnerability to natural occurrences. It is devoid of historical experiences, politics, power dynamics, and cultural elements. Technology development for intricate livelihood strategies can be aided by integrating the framework with other conceptual frameworks (Adato and Meinzen-Dick, 2002)

studies have shown the microorganisms in ozonated soil can improve pre-ozonation and bioremediation techniques. After 0-900 minutes of ozonation, diesel-contaminated soils showed a notable decrease in total PH and aromatics. During the nine weeks of bioremediation, the number of microorganisms increased during incubation. Significant removal was noted in 900-minute soil; however, the maximum removal (25.4%) was found in 180-minute ozonated soil. (Ahn et al., 2005)

Baheri and Meysami studied the use of fungal bioaugmentation in lab-scale composters to break down flare pit dirt. TPH (total petroleum hydrocarbon) was 16% at first. The TPH concentration of the soil decreased by 29% after 98 days, and the oil extract demonstrated a 70–99% reduction. Statistical research, however, did not reveal any noteworthy effects from the application of fungi or moisture content. The amount of bulking agent has a negligible impact on hydrocarbon loss (Baheri, and Meysami 2002).

Balkema et al. they studied decentralized wastewater treatment systems for emission prevention is discussed in the study, with an emphasis on efficient use of resources. In addition to offering a basic evaluation methodology based on multi-objective optimisation and sustainability indicators, it includes a survey of the literature on sustainability assessment techniques and indicators. This method assists in weighing the trade-offs while choosing environmentally friendly wastewater treatment solutions (Balkema et al. 2002)

2. Methodology

2.1. *Microorganism and inoculum preparation*

Process designing was used to the nearby CO_2 liberal microalga strain Desmodesmus pannonicus CT01 to increase intracellular protein content and advancement execution according to (Beyers et al., 2001). Each assessment's inoculum was filled in Basal Bold's medium (BBM), which had a major pH of 8 (Bidoki et al., 2006). The cultures were made in 250 mL Erlenmeyer flasks with a 100 mL working volume (BIO-PRO., 2008). They

were struggled in a shaker hatching focus with tumult at 150 rpm and room temperature (Bitton, 2005).

2.2. *Media optimization for maximization of biomass titer and productivity.*

The design comprises twenty exploratory runs at three coded levels, with five repetitions at the Centre highlight to quantify the trial mistake the variables' high, medium, and low levels were denoted by the values +α, +1 for high, 0 for medium, and -1, −α for low as shown in table 1.

Table 1: To maximize biomass titer and production, the chosen Basal Bold's media components' actual and coded levels

Factors			Levels and corresponding actual values			
Code	**Media Components**	**Units**	**-1 Level**	**+1 Level**	**- alpha**	**+alpha**
A	Initial KH2PO4 concentration	mg L-1	130.40	219.60	100	250
B	Initial NaNO3 concentration	g L-1	0.45	1.05	0.25	1.25
C	Initial TME units	units	0.40	0.85	0.25	1

Twenty starter combinations, including eight factorial points, six centre points, and six duplicates of the centre points, were expected by the CCD. To make sense of the shared collaboration between the variables and their associated ideal levels, a second-request polynomial condition was used. This condition can be expressed overall as follows:

The exploratory data from the CCD was bankrupt down using various regression analysis to distinguish significant changes ($p < 0.05$) in responses under various settings. To perceive the best conditions and visualize the correlations between the responses and the autonomous variables, 3D surface plots were made (Cole-Turner, 2003; Blanco, 2000; Blonskaya and Vaalu, 2006) as shown in table 2

Table 2: Experimental and predicted biomass production and productivity data under varying conditions.

				Experimented		Predicted	
Std. No.	**KH2PO 4 (mg L-1)**	**NaNO3 (g L-1)**	**TME (units)**	**Biomas s (g L-1)**	**Productivity (mg L-1 day-1)**	**Biomas s (g L-1)**	**Productivity (mg L-1 day-1)**
1	130.405	0.4527	0.40202	0.6625	20.3	0.66	20.21
2	219.595	0.4527	0.40202	0.661	22.45	0.66	22.41
3	130.405	1.0473	0.40202	0.6879	25.81	0.69	25.77
4	219.595	1.0473	0.40202	0.7812	23.78	0.78	23.84
5	130.405	0.4527	0.84798	0.6818	20.61	0.68	20.56
6	219.595	0.4527	0.84798	0.7539	28.39	0.75	28.45
7	130.405	1.0473	0.84798	0.7821	24.15	0.78	24.21
8	219.595	1.0473	0.84798	0.9512	27.87	0.95	27.97
9	100	0.75	0.625	0.6415	18.41	0.64	18.49
10	250	0.75	0.625	0.786	23.6	0.79	23.5

11	175	0.25	0.625	0.6516	22.16	0.65	22.24
12	175	1.25	0.625	0.8412	26.61	0.84	26.51
13	175	0.75	0.25	0.7299	26.47	0.73	26.54
14	175	0.75	1	0.8878	30.4	0.89	30.31
15	175	0.75	0.625	0.9555	31.66	0.96	31.59
16	175	0.75	0.625	0.9562	31.61	0.96	31.59
17	175	0.75	0.625	0.9553	31.58	0.96	31.59
18	175	0.75	0.625	0.9571	31.63	0.96	31.59
19	175	0.75	0.625	0.9567	31.52	0.96	31.59
20	175	0.75	0.625	0.9577	31.51	0.96	31.59

2.3. *Influence of light intensity and wavelength on the rate of development and metabolic makeup of CT01*

A 500 mL working volume, layer sparger-equipped, specifically developed barrel-shaped film bubble segment transport photobioreactor (CM-APBR) was utilized for the lifestyle (Commission of the European Communities. 2002) as shown in figure 1. It was made of a 75 mm outer diameter, 69 mm inner diameter, and 175 mm height acrylic tube with flanges. For the purpose of film sparger, a neoprene elastic sheet with a thickness of 2 mm was utilised. The reactor's surface (one side) was aligned with four Drove panels, each measuring 3 x 50 W, so that individual wavelengths could be studied (Das and Mukherjee, 2007; Dale and Kim, 2006). A consistent light intensity of 100 µE m-2 s-1 was present on all of the boards. Two more Driven panels, with a ratio of 1:2 and maintaining the same intensity, were inserted from different sides of the reactor to explore mixed wavelengths. A DC voltage regulator is attached to every Drove board, allowing you to modify the light intensity as needed. Following that, the optimal combination of light wavelengths was used to evaluate the biochemical composition and improvement performance of the CT01 strain at four distinct light intensities (50, 100, 150, and 200 µE m-2 s-1). The improved BBM medium was used for all studies, with a light: dark pattern of 16:8 hours and an air stream pace of 0.5 vvm. Normal sampling intervals were used to follow the biomass' development and composite (Das, 2005; Das and Jain, 2001).

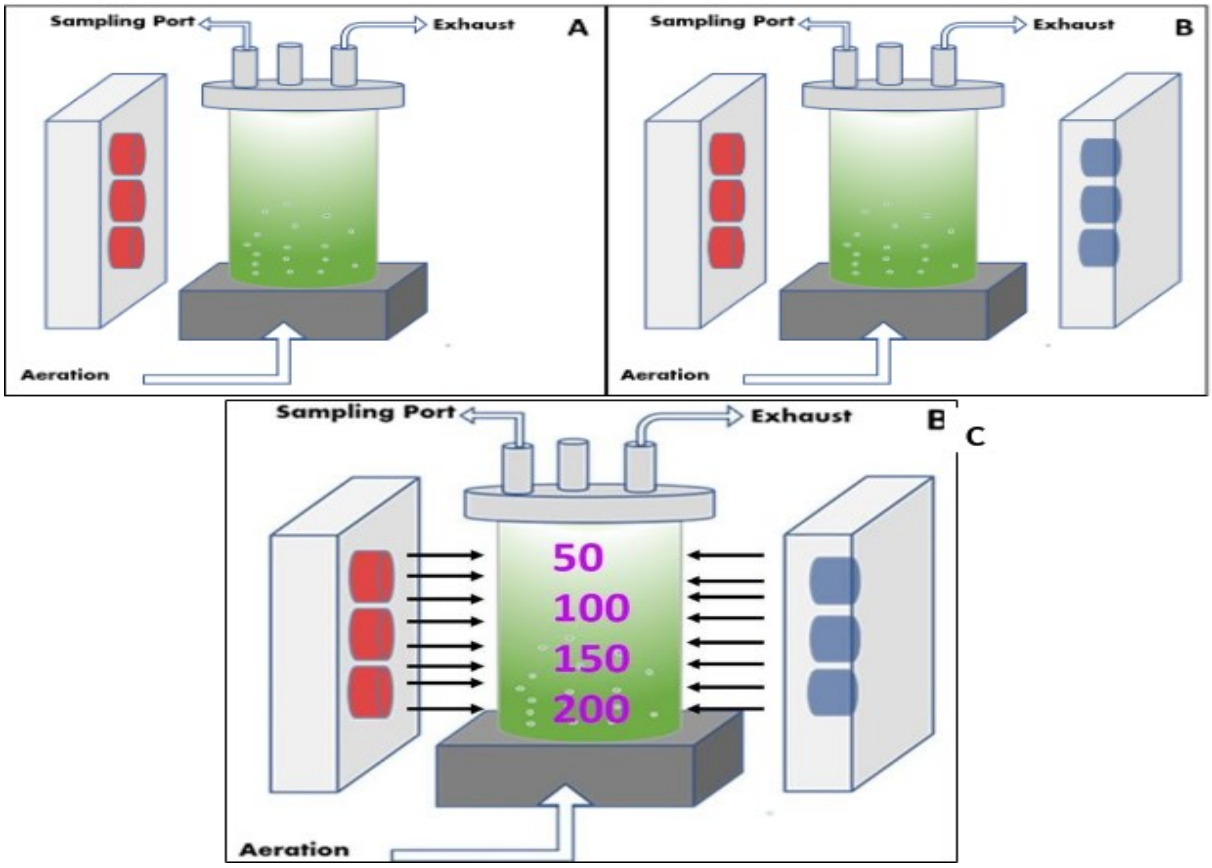

Figure 1: Diagrammatic representation of the micro reactor's experimental setup: Three conditions for culture exist: (A) monochromatic light, (B) mixed light, and (C) varying light intensity

2.4. *Encouraging CT01 growth using pH-based CO_2 feeding*

To additional increase biomass titer and efficiency, media components were advanced and a brightening strategy was created (Dash, 2009). Then, a process designing strategy based on CO_2 benefiting from demand was used to keep the way of life pH inside the enhanced reach. The microreactor used to develop the way of life was equipped with an upgraded light frequency of 100 µE m-2 s-1 intensity, enhanced BBM medium, and an ideal air circulation pace of 0.5 vvm. Using cascade control and a solenoid valve to convey CO_2 to the way of life, the pH of the way of life was kept inside its ideal scope of 8 to 8.5 during the entire development process, all under the bearing of the pH sensor (De Souza et al., 2022; Charpentier, 2007). There were two batches run: one with pH control and the other without a pH control cascade (De Steur et al., 2015). All through the cluster's span, sampling was finished on a regular basis to procure a unique development profile, and the way of life pH was measured like clockwork. At the point when stationary phase started, the clump was stopped (Falck-Zepeda et al., 2000; Eisenhut and Weber, 2019).

2.5. *Examination of development, biomass make-up, and substrate application*

Optical density at 600 nm was used as an index to analyze the growth of cells with spectrophotometer (Gonsalves et al., 2004). The kinetic growth data obtained by monitoring cell concentration was fitted to the Monod equation (Hines et al., 2021). Phosphate and nitrate consumption was

measured by colorimetric method and ion chromatography (Iñiguez et al., 2021). The contents of sugars, proteins and lipids in the dry weight were determined following standard biochemical methods. The analysis of sugar was based on phenol-sulfuric acid method. The proteins were quantified using Bradford assay. The quantification of lipids was implemented by extraction with chloroform-methanol mixture and the performed gravimetric determination. Chlorophyll extraction. The cell pellets were incubated in 90% methanol at 45°C for 30 min. An aliquot of the methanolic extracts were separated from cell debris by a centrifugation of 10, 000 g for 10 min at 4°C. Chlorophyll content was determined using spectrophotometry (Kromdijk et al., 2016).

3. **Results and Discussions**

3.1. *Maximising biomass titre and production by statistical optimisation of CT01 medium*

By raising the union of relevant elements utilising the RSM CCD, we significantly improved CT01's biomass obsession and efficacy this observation is in agreement with the result of (Murchie et al., 2015) Additionally, the interplay between several nutrients, such as potassium dihydrogen phosphate, sodium nitrate, and trace and microelements (TME), and their effects on enhanced metrics was deduced. All exploratory data were fitted using second request polynomials after data was separated using standard analysis of variance (ANOVA) as show in table 3.

A represents the phosphate content, B the nitrate concentration, and C the total dissolved solids (TME) concentration, and Y the normal response as shown in figure 2.

Table 3: ANOVA for the CCD-derived quadratic regression model used to optimise the medium components for biomass productivity

Source	Sum of Squares	df	Mean Square	F-Value	P-Value prob > F	
MODEL	0.4	9	0.144	19231.78	< 0.0001	Significant
A-KH2PO4	0.135	1	0.135	14233.92	< 0.0001	Significant
B-NaNO3	0.157	1	0.154	24829.40	< 0.0001	Significant
C-TME	0.13	1	0.14	17652.32	< 0.0001	Significant
A2	4.7E-14	1	4.71E-14	2684.17	< 0.0001	Significant
B2	2.80E-14	1	2.80E-14	1632.97	< 0.0001	Significant
C2	2.90E-14	1	2.90E-14	1689.9	< 0.0001	Significant
AB	0.22	1	0.22	61570.36	< 0.0001	Significant
AC	0.180	1	0.180	45935.15	< 0.0001	Significant
BC	0.140	1	0.140	22729.25	< 0.0001	Significant
Residual Error	1.83E-16	10	1.83E-17			
Lack-of-Fit	1.30E-16	5	2.70E-28	2.97	0.1284	Not significant

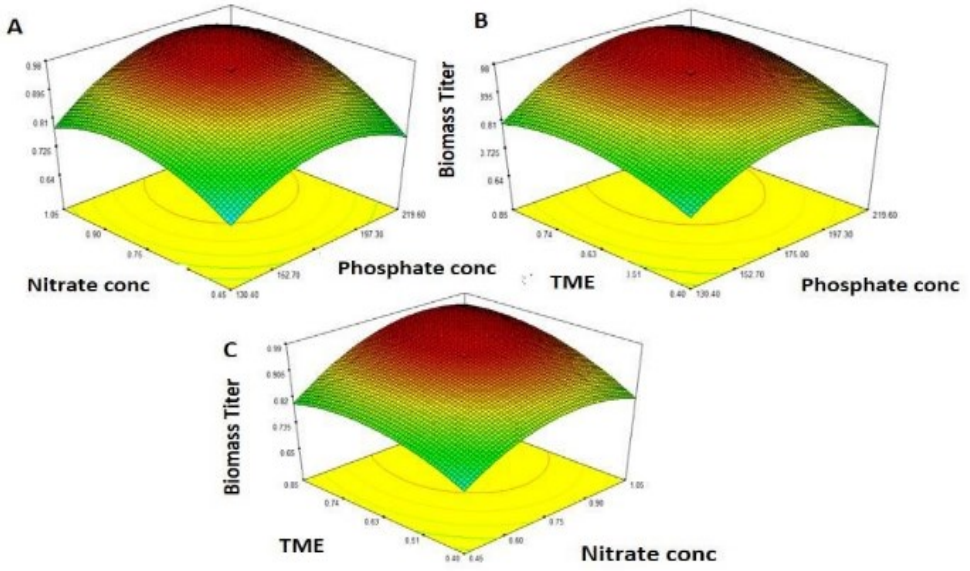

Figure 2: Plots showing response surfaces that show how phosphate and nitrate, TME and phosphate, and TME and nitrate interface with biomass titer. The third factor was kept up with at its center worth during the communication of the first two variables. Phosphate, nitrate, and TME had center values of 0.175 g L-1, 0.75 g L-1, and 0.63 units, respectively.

It was demonstrated that the models resolved on the importance of the preliminary data since the ANOVA of the turn of events and efficacy quadratic regression models indicated prob. There was no statistically significant lack of fit for any answer, and the model was confirmed to be fit by the coefficient of assurance (R2). The R2 value for efficiency was 0.9997 and for biomass enhancement it was 0.9999.

The variables that were determined to have a substantial impact on biomass yield were examined using response surface plots. The three components interact with each other to maintain one variable at an ideal level, which in turn affects efficiency and biomass production. Using RSM design to further refine the medium focus, we were able to create a compelling medium with adjusted supplement content, which led to increased productivity and the most absurd biomass creation. The model was probably evaluated using the distinguished ideal conditions. Previous studies on *S. vacuolatus* have shown similar results, highlighting the need to refine nitrate and phosphate concentrations for microalgal enhancement. There was a marked improvement with increasing phosphate and nitrate contents. Numerous studies have shown that microalgal species development can be affected by low nitrate or phosphate concentrations, making these nutrients crucial for development and production control.. A 26.67% increase in productivity compared to the unoptimized condition and a 28.3% increase in biomass compared to the unoptimized medium (0.7541 g L-1) provided evidence in favour of the statistical technique that was put into action, this observation is in agreement with the result of (Ogbaga et al., 2015; Ruangsomboon et al., 2007) .

3.2. Interaction between light intensity and wavelength for the purpose of producing biomass with a high protein density

Further developing both the biomass' intracellular protein content and development execution is essential to making the strain CT01 as a substitute for conventional aqua feed. To accomplish our objective, we exposed the CT01 strain to various light wavelengths and intensities. This conclusion was consistent with findings from previous research that demonstrated the superior exhibition of a blend of red and blue light wavelengths. Among mono-wavelength, it has been noticed that red light's developing execution as shown in figure 3 and 4.

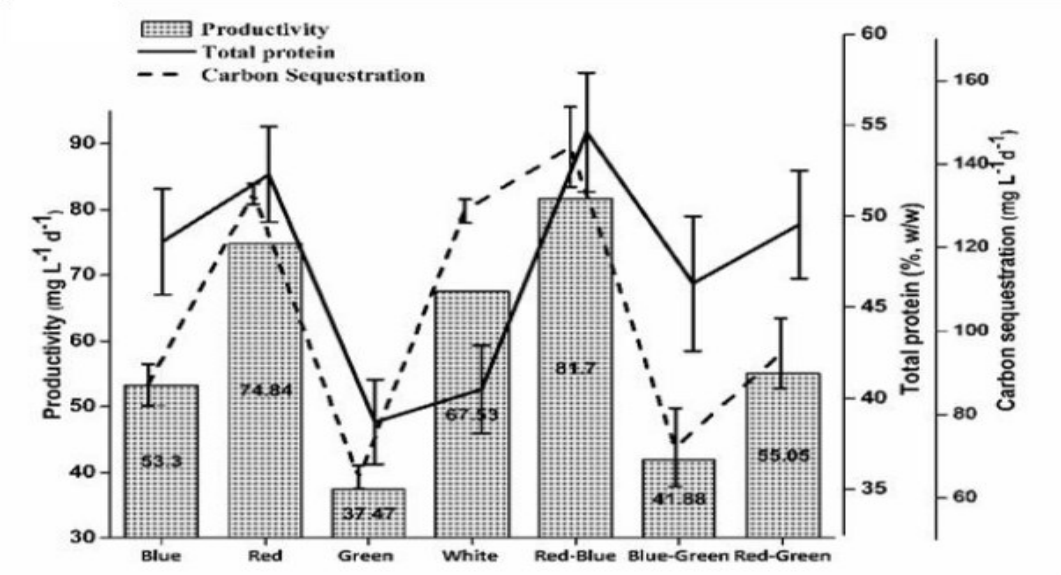

Figure 3: Carbon sequestration rate, total protein content, and biomass productivity of Desmodesmus pannonicus CT01 subjected to various light wavelengths

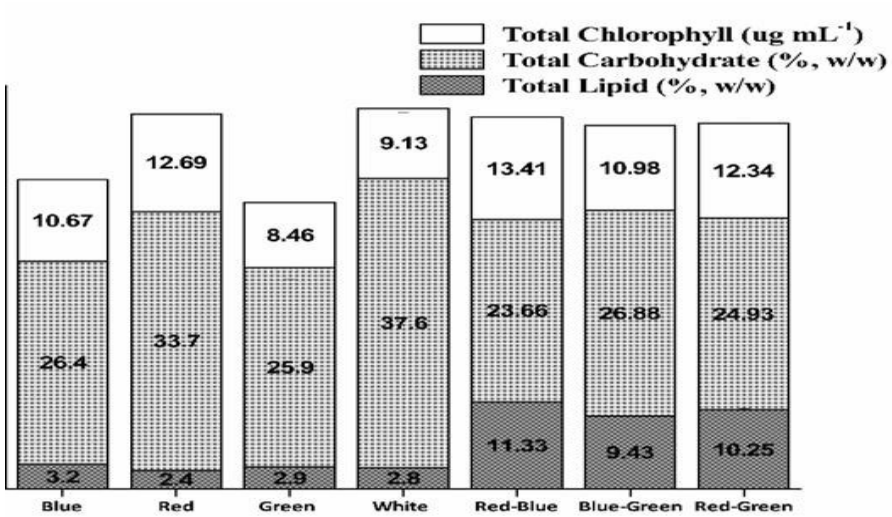

Figure 4: Variations in light frequency impact on the complete chlorophyll, absolute sugar, and all out-lipid content of Desmodesmus pannonicus CT01 biomass.

It was discovered that CT01's growth performance was lower when green wavelengths, or combinations of red-green or blue-green wavelengths, were present this observation is in agreement with result of (Parmar et al., 2017). Comparable patterns were observed over the whole range of light wavelengths, since the carbon sequestration rate and CT01 cell productivity are correlated. These results may be connected to a study that found that *Chlorella sp*. grown in blue and red LED environments had greater biomass concentrations, productivity levels, and specific growth rates than those grown in white and green LED environments. Various microalgal species exhibit diversity in their adoption of light spectrum, as previously reported. Therefore, optimizing illumination becomes imperative. It has previously been observed that for microalgae and plants to engage in effective photosynthesis, red and blue light must be supplied in suitable amounts as shown in figure 5.

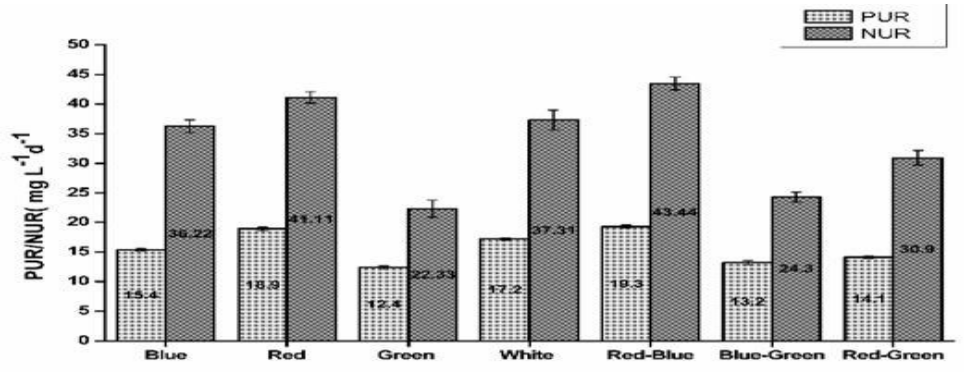

Figure 5: Desmodesmus pannonicus CT01 biomass's nutrient utilisation rate profile when exposed to various light wavelengths.

This may generally improve nitrogen metabolism, light-harvesting complexes, and chlorophyll production. There are several environmental factors that can affect how microalgae develop, but one of the most important ones is the amount of light that strikes the cells. Light is the single most important need for photosynthetic activity and is also necessary for microalgal autotrophic growth. In addition to respiration and photosynthesis, it plays a part in cell division. In order to produce ATP and NADPH and to synthesise other critical molecules for the development of microalgae, light is necessary.

Microalgae come in a wide variety of species, and each one has a preferred light intensity for development and biomass production. Other factors, such the temperature and the presence of nutrients in the growth media, also affect this ideal light intensity. It is more likely to produce more biomass the more intense the light. This is due to the photosynthetic apparatus's increased capacity for light absorption and utilization as shown in figure 6.

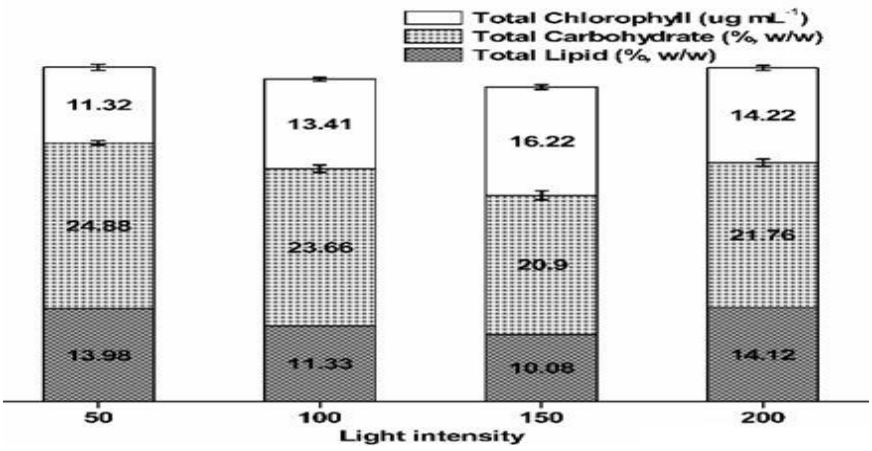

Figure 6: Contents of total chlorophyll, carbohydrates, and fats in Desmodesmus pannonicus CT01 biomass under varying light intensity exposure.

The specific kind of green growth and the development climate influence this saturation threshold. In the ongoing investigation, four distinct red-blue light intensities were applied to the CT01 strain in a 1:1 proportion. In an associated investigation, when the light intensity from blue Drove lamps rose from 100 to 200 µE m-2 s-1, *C. vulgaris* fostered even more successfully. This finding might be associated with a study that discovered that complete protein content increased as irradiance increased. As indicated (Ogbonda et al. 2007) Spirulina sp. may deliver more biomass and go through more protein biosynthesis in response to increased irradiance. Supplement use rose directly as light intensity increased and started to decline at 200 µE m-2 s-1. This can be explained by the way that the multiplication of microalgal cells and the usage of macronutrients are associated. Numerous studies demonstrate that when irradiance increases, phosphate and nitrate admission increase as well, prompting an increase in cell density this observation is in agreement with (Thieman, and Palladino, 2008) as shown in figure 7.

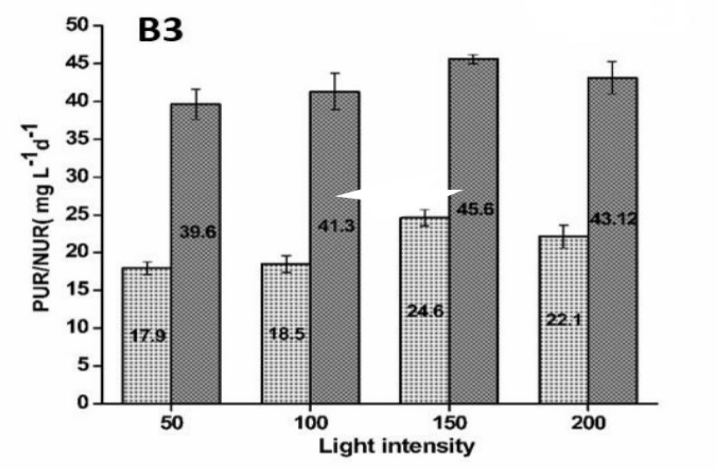

Figure 7: Desmodesmus pannonicus CT01 biomass's nutrient utilisation rate profile under various light intensity exposures

3.3. Results of CT01's growth when fed CO_2 according to pH

With regards to upgrading phototrophic development in microalgae, observing pH levels in the way of life provides roundabout insight into the accessibility of CO_2 to the organisms. This involves understanding the distribution of dissolved inorganic carbon species, such as aqueous carbon dioxide, bicarbonate, and carbonate, which change with pH levels. To improve CO_2 usage and defeat carbon source limitations, a pH-based CO_2 was carried out for the CT01 strain, expanding upon previous research. By incorporating this strategy with ideal culture conditions and a devised enlightenment plan, two distinct batches were contrasted — one and pH control and one without. Results showed that the pH-controlled group displayed superior biomass effectiveness and carbon sequestration rate, featuring the adequacy of CO_2 supplementation. Supplement usage rates also increased significantly in the pH-controlled group, showing great conditions for CT01 multiplication and further developed development and creation. Surprisingly, the biochemical composition of CT01 remained to a great extent consistent across the two batches, emphasizing the utility of pH-based CO_2 control in improving microalgae development without modifying biomass composition this observation is in agreement with (Wei et al.,2022; Wu et al., 2019; Zuo et al., 2018) as shown in figure 8,9,10 and 11.

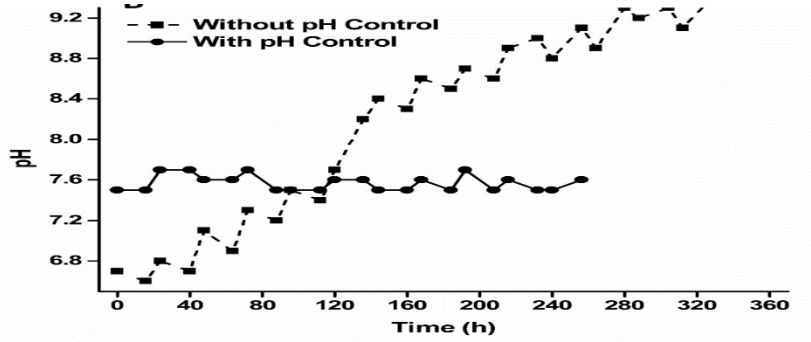

Figure 8: Collaborative patterns of CT01 growth when cultivated with or without pH control, in conjunction with the well-established process engineering method for light intensity and wavelength

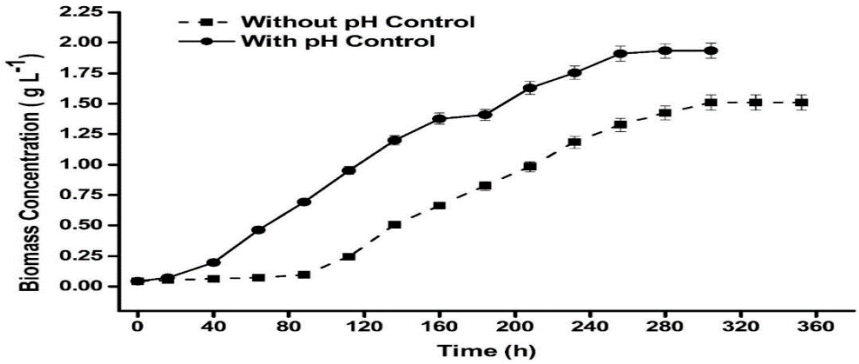

Figure 9: Joining the unique pH profiles of the stock with the previously described process designing strategy for light frequency and intensity during CT01 development with and without pH control

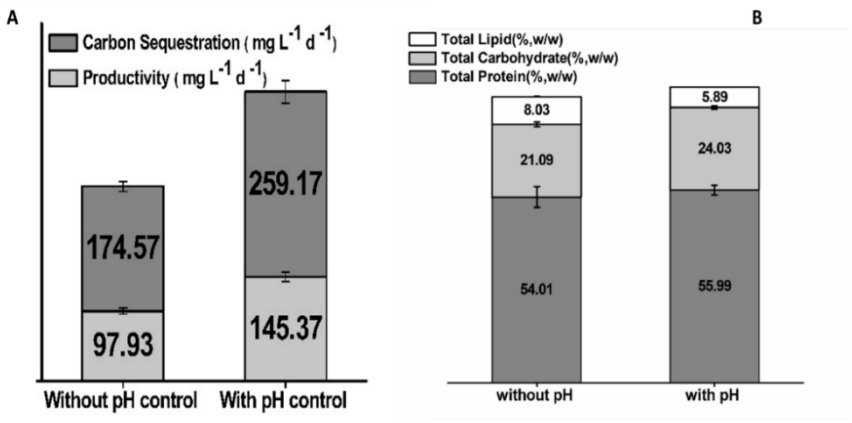

Figure 10: (A) The speed of carbon sequestration and generally efficiency; (B) the biomass composition of CT01 batches become both with and without pH control using the previously established process designing strategy for light intensity and repeat

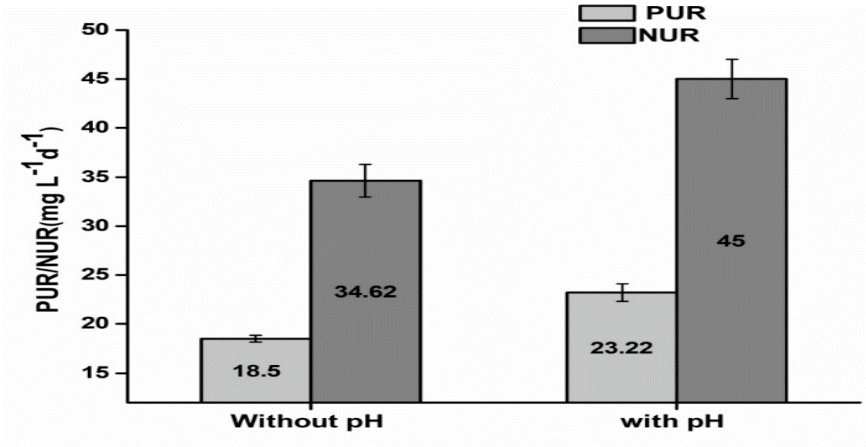

Figure 11: Supplement use rate profile of CT01 batches developed using the previously established process designing strategy for light intensity and frequency the executives and without pH control.

4. Conclusions

The CCD approach was utilized to enhance the concentrations of sodium nitrate, dihydrogen potassium phosphate, and full microbial concentrate (TME) in BB media to increase biomass quantity and efficacy. This optimization resulted in a 28% increase in biomass fixation and a 27% increase in biomass production compared to unoptimized conditions, demonstrating significant improvements in process variables through the use of CCD-RSM. To further enhance intracellular biomass components and development characteristics, the CT01 strain was exposed to seven distinct combinations of single and multiple recurrent light exposures after establishing the BB medium. The best results for CT01 were achieved with a combination of red and blue light frequencies, significantly boosting biomass production, carbon sequestration rate, and total protein content. Subsequently, CT01 was subjected to varying light intensities while maintaining the optimal medium and red-blue light combination. At an intensity of 150 µE m-2 s-1, the biomass efficiency and carbon sequestration rate of the CT01 strain significantly improved, although only a slight increase in protein content was observed. A process design strategy incorporating the on-demand supply of CO_2 under variable light intensities could enhance overall effectiveness and biomass concentrations by eliminating CO_2 limitation and maintaining optimal culture pH, given the significant interplay between optimization, culture pH, and light intensity. The group with uncontrolled pH produced the lowest biomass concentrations (1.93 g L-1), whereas the group with controlled pH showed a 28% improvement and consumed the most phosphate and supplements. This method appears to enhance supplement consumption, suggesting that wastewater treatment sites could derive additional benefits from its implementation.

References

Adato, M., & Meinzen-Dick, R. (2002). Assessing the impact of agricultural research on poverty using the Sustainable Livelihoods Framework. International Food Policy Research Institute.

Ahn, Y., Jung, H., Tatavarty, R., Choi, H., Yang, J., & Kim, I. S. (2005). Monitoring of petroleum hydrocarbon degradative potential of indigenous microorganisms in ozonated soil. Biodegradation, 16(1), 45-56.

Ah-You, K., Suleiman, M., & Jaworski, J. (2000). Biotechnology and cleaner production in Canada. Program for Energy Research and Development (PERD), Life Sciences Branch Industry Canada.

Alivisatos, P. (2004). The use of nanocrystals in biological detection. Nature Biotechnology, 22(1), 47-52.

Alkhaddar, R. M., Phipps, D. A., & Cheng, C. (2005). Today and tomorrow! Research prospects for aerobic biological liquid waste treatment for reduction of carbon load. E-Water, Official Publication of the European Water Association (EWA), 1-18.

Andres, Y., Dumont, E., Le Cloirec, P., & Ramirez-Lopez, E. (2006). Wood bark as packing material in a biofilter used for air treatment. Environmental Technology, 27(12), 1297-1301.

Baheri, H., & Meysami, P. (2002). Feasibility of fungi bioaugmentation in composting a flare pit soil. Journal of Hazardous Materials, 89(2-3), 279-286.

Balkema, A. J., Preisig, H. A., Otterpohl, R., & Lambert, F. (2002). Indicators for the sustainability assessment of wastewater treatment systems. Urban Water, 4(2), 153-161.

Baptista, S. J., Camporese, E. F. S., & Freire, D. D. C. (2006). Evaluation of biostimulation, bioaugmentation and use of biosurfactant as treatment technique of clay soil contaminated with diesel oil. Environmental Engineering and Management Journal, 6(2), 1325-1332.

Beaudette, L. A., Cassidy, M. B., England, L., Kirk, J.L., Habash, M., Lee, H., & Trevors, J. T. (2002). Bioremediation of soils. In G. Bitton (Ed.), Encyclopedia of Environmental Microbiology, 722-737. Wiley-Interscience.

Beck-Friis, B. G. (2001). Emissions of ammonia, nitrous oxide and methane during composting of organic household waste (PhD thesis). Swedish University of Agricultural Sciences, Uppsala.

Beliaeff, B., & Burgeot, T. (2002). Integrated biomarker response: A useful tool for ecological risk assessment. Environmental Toxicology and Chemistry, 21(6), 1316-1322.

Beyers, L., Ismaël, Y., Piesse, J., & Thirtle, C. (2001, June). The efficiency of Bt cotton adopters in the Makhathini Flats of Kwazulu-Natal. Paper presented at the ISNAR consultation "Biotechnology and Rural Livelihood—Enhancing the Benefits," The Hague.

Bidoki, S. M., Wittlinger, R., Alamdar, A. A., & Burger, J. (2006). Eco-efficiency analysis of textile coating materials. Journal of the Iranian Chemical Society, 3(4), 351-359.

BIO-PRO. (2008). Biotechnology in the chemical industry: The long road from exception to standard (II). The Biotech/Life Science Portal Baden-Württemberg. Retrieved from http://bio-pro.de/en/region/ulm/magazin/00698/index-html

Bitton, G. (2005). *Wastewater microbiology*. Wiley-Liss, John Wiley and Sons.

Blanco, A. (2000). Immobilization of non-viable cyanobacteria and their use for heavy metal adsorption from water. In E. J. Olguin, G. Sánchez, & E. Hernández (Eds.), *Environmental biotechnology* (pp. 135-151). Taylor & Francis.

Blonskaya, V., & Vaalu, T. (2006). Investigation of different schemes for anaerobic treatment of food industry wastes in Estonia. *Proceedings of the Estonian Academy of Sciences: Chemistry, 55*, 14-28.

Cole-Turner, R. (2003). Biotechnology. In Encyclopedia of Science and Religion. Retrieved December 7, 2014, from https://www.encyclopedia.com/science/encyclop

edias-almanacs-transcripts-and-maps/biotechnology

Commission of the European Communities. (2002). Economic impacts of genetically modified crops on the agrifood sector: A first review. Working Document Rev. Directorate General for Agriculture.

Charpentier, J.C. (2007). In the frame of globalization and sustainability, process intensification, a path to the future of chemical and process engineering (molecules into money). Chemical Engineering Journal, 134(1-3), 84-92.

Dale, B. E., & Kim, S. (2006). Biomass refining global impact – The biobased economy of the 21st century. In B. Kamm, P. R. Gruber, & M. Kamm (Eds.), Biorefineries - Industrial processes and products. Status quo and future directions, 1 ,41-66. Wiley-VCH.

Das, K., & Mukherjee, A. K. (2007). Crude petroleum-oil biodegradation efficiency of *Bacillus subtilis* and *Pseudomonas aeruginosa* strains isolated from a petroleum-oil contaminated soil from North-East India. *Bioresource Technology, 98*(7), 1339-1345.

Das, T. K. (2005). Toward zero discharge: Innovative methodology and technologies for process pollution prevention. John Wiley and Sons.

Das, T. K., & Jain, A. K. (2001). Pollution prevention advances in pulp and paper processing. Environmental Progress & Sustainable Energy, 20(2), 87-92.

Dash, R. R., Gaur, A., & Balomajumder, C. (2009). Cyanide in industrial wastewater and its removal: A review on biotreatment. Journal of Hazardous Materials, 163(1), 1-11.

De Souza, A. P., Burgess, S. J., Doran, L., Hansen, J., Manukyan, L., Maryn, N., (2022). Soybean photosynthesis and crop yield are improved by accelerating recovery from photoprotection. Science, 377(6608), 851-854. https://doi.org/10.1126/science.adc9831

De Steur, H., Blancquaert, D., Strobbe, S., Lambert, W., Gellynck, X., & Van Der Straeten, D. (2015). Status and market potential of transgenic biofortified crops. Nature Biotechnology, 33(1), 25-29. https://doi.org/10.1038/nbt.3110

Eisenhut, M., & Weber, A. P. M. (2019). Improving crop yield. Science, 363(6422), 32-33. https://doi.org/10.1126/science.aav8979

Falck-Zepeda, J. B., Traxler, G., & Nelson, R. G. (2000). Surplus distribution from the introduction of a biotechnology innovation. American Journal of Agricultural Economics, 82(2), 360-369.

Gonsalves, C., Lee, D. R., & Gonsalves, D. (2004). Transgenic virus-resistant papaya: The Hawaiian "Rainbow" was rapidly adopted by farmers and is of major importance in Hawaii today. APSnet Feature Articles. https://doi.org/10.1094/APSnetFeature-2004-0804

Hines, K. M., Chaudhari, V., Edgeworth, K. N., Owens, T. G., & Hanson, M. R. (2021). Absence of carbonic anhydrase in chloroplasts affects C3 plant development but not photosynthesis. Proceedings of the National Academy of Sciences, 118(18), e2107425118. https://doi.org/10.1073/pnas.2107425118

Iñiguez, C., Aguiló-Nicolau, P., & Galmés, J. (2021). Improving photosynthesis through the enhancement of Rubisco carboxylation capacity. Biochemical Society Transactions, 49(5), 2007-2019. https://doi.org/10.1042/BST20201056

Kromdijk, J., Głowacka, K., Leonelli, L., Gabilly, S. T., Iwai, M., Niyogi, K. K., (2016). Improving photosynthesis and crop productivity by accelerating recovery from photoprotection. Science, 354(6314), 857-861. https://doi.org/10.1126/science.aai8878

Murchie, E. H., Ali, A., & Herman, T. (2015). Photoprotection as a trait for rice yield improvement: Status and prospects. Rice, 8(1), 31. https://doi.org/10.1186/s12284-015-0065-2

Ogbaga, C. C., Stepien, P., Athar, H.-U.-R., & Ashraf, M. (2018). Engineering Rubisco activase from thermophilic cyanobacteria into high-temperature sensitive plants. Critical Reviews in Biotechnology, 38(4), 559-572. https://doi.org/10.1080/07388551.2017.1378998

Ogbonda, K. H., Abu, G. O., & Aminigo, R. E. (2007). Optimization studies of biomass production and protein biosynthesis in a Spirulina sp. isolated from an oil-polluted flame pit in the Niger Delta. African Journal of Biotechnology, 6(22), 2550-2554.

Parmar, N., Singh, K. H., Sharma, D., Singh, L., Kumar, P., Nanjundan, J., (2017). Genetic engineering strategies for biotic and abiotic stress tolerance and quality enhancement in horticultural crops: A comprehensive review. 3 Biotech, 7(4), 239. https://doi.org/10.1007/s13205-017-0870-y

Ruangsomboon, S., Chidthaisong, A., Bunnag, B., Inthorn, D. and Harvey, N.W. (2007). Lead (Pb2+) adsorption characteristics and sugar composition of capsular polysaccharides of cyanobacterium Calothrix marchica Songklanakarin J. Sci. Technol., 29(2), 529-541

Thieman, W. J., & Palladino, M. A. (2008). Introduction to biotechnology. Pearson/Benjamin Cummings.

Wei, S., Li, X., Lu, Z., Zhang, H., Ye, X., Zhou, Y., (2022). A transcriptional regulator that boosts grain yields and shortens the growth duration of rice. Science, 377(6608), eabi8455. https://doi.org/10.1126/science.abi8455

Wu, J., Lawit, S. J., Weers, B., Sun, J., Mongar, N., Van Hemert, J., (2019). Overexpression of zmm28 increases maize grain yield in the field. Proceedings of the National Academy of Sciences, 116(48), 23850-23858. https://doi.org/10.1073/pnas.1902593116

Zuo, L., Yang, R., Zhen, Z., Liu, J., Huang, L., & Yang, M. (2018). A 5-year field study showed no apparent effect of the Bt transgenic 741 poplar on the arthropod community and soil bacterial diversity. Scientific Reports, 8(1), 1956. https://doi.org/10.1038/s41598-018-20322-3