



Biotechnological Optimisation of *Spirulina platensis* Cultivation for Enhanced Antibacterial Activity and Commercial Applications

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Abstract

Spirulina platensis is a filamentous, alkaliphilic cyanobacterium of substantial nutritional and pharmaceutical value, and there is growing interest in biotechnological strategies that enhance its bioactive potential for commercial exploitation. The present study investigated the optimization of three principal physicochemical cultivation parameters, namely initial medium pH, light intensity, and incubation temperature, with a view to enhancing biomass yield and antibacterial activity, and evaluated the resulting biomass for commercial applicability. Using a one-factor-at-a-time approach, the optimum conditions were identified as pH 9.5, light intensity 2000 lux, and temperature 32 °C, under which dry biomass increased by 65.7% relative to the control (1.74 versus 1.05 g L⁻¹). Extracts prepared from optimized biomass using solvents of differing polarity were evaluated for antibacterial activity by the agar well diffusion assay, minimum inhibitory concentration (MIC), and minimum bactericidal concentration (MBC) against eight Gram-positive and Gram-negative bacteria. The methanolic extract exhibited the greatest broad-spectrum, predominantly bactericidal activity, with zones of inhibition ranging from 12.9 to 18.6 mm and MIC values of 62.5–156.0 µg mL⁻¹. Bioactive constituents, including phycocyanin, chlorophyll a, carotenoids, phenolics, and flavonoids, increased by 44.9–51.9% under optimized conditions, and a strong positive correlation ($r = 0.94$) was observed between biomass yield and antibacterial activity. Commercial feasibility assessment indicated strong potential across the pharmaceutical, nutraceutical, cosmetic, food-preservation, and natural-antimicrobial sectors. These findings demonstrate that the rational manipulation of readily controllable cultivation parameters constitutes an effective, scalable biotechnological strategy for producing antibacterially enhanced *S. platensis* biomass of commercial value.

Keywords: *Spirulina platensis*; cultivation optimization; antibacterial activity; biotechnology; commercial application

1. Introduction

1.1 Background of *Spirulina platensis* and Its Industrial Significance

Spirulina platensis, currently reclassified within the genus *Arthrospira*/*Limnospira*, is a multicellular, helically coiled, filamentous cyanobacterium that thrives naturally in alkaline lakes and has been consumed as a food source for centuries (Spínola et al., 2024). Its industrial cultivation has expanded considerably over the past two decades owing to its exceptionally high protein content, rich pigment profile, and favourable nutritional composition, which have secured its position as one of the most commercially significant

microalgae worldwide (Sinetova et al., 2024). Beyond its established role as a dietary supplement, *S. platensis* biomass is increasingly recognized as a versatile biotechnological feedstock capable of yielding pharmaceutically and industrially relevant bioactive compounds.

The organism's amenability to large-scale cultivation in open raceway ponds and closed photobioreactors, its rapid doubling time under favourable conditions, and its long history of safe human consumption together distinguish it from many other microalgae as a commercially tractable production platform (Grosshagauer et al., 2020). These attributes, combined with a growing global market for natural, plant- and microbe-derived functional ingredients, have created strong commercial incentives for research directed at improving the productivity and functional quality of cultivated *S. platensis* biomass.

1.2 Antibacterial Potential of Cyanobacterial Biomolecules

Cyanobacteria such as *S. platensis* synthesize a diverse array of secondary metabolites, including phycobiliproteins, phenolic acids, flavonoids, and polyunsaturated fatty acids, several of which possess demonstrable antibacterial activity against both Gram-positive and Gram-negative pathogens (Usharani et al., 2015; Ilieva et al., 2024). The growing global concern surrounding antibiotic resistance has intensified interest in natural, multi-target antimicrobial agents of this kind, since their multi-constituent mode of action is considered less liable to elicit resistance than single-target synthetic antibiotics (Al-ghanayem, 2017). The extraction and characterization of these antibacterial constituents from *S. platensis* therefore represents a research direction of considerable applied importance.

Reported antibacterial mechanisms attributed to *Spirulina*-derived constituents include disruption of bacterial membrane integrity by phenolic and pigment compounds, interference with microbial enzyme systems, and direct peptide-mediated bactericidal action (Sun et al., 2016; Gheda & Ismail, 2020). Because these mechanisms act through several independent routes rather than a single molecular target, extracts of this kind are generally regarded as promising candidates for applications in which the emergence of resistance is a significant concern, including topical antiseptics and food preservation.

1.3 Role of Physicochemical Parameters in Cultivation Optimization

The growth, biochemical composition, and bioactivity of *S. platensis* are strongly influenced by the physicochemical conditions of cultivation, particularly the pH of the medium, the intensity of illumination, and the incubation temperature (Zhai et al., 2017). Because *S. platensis* is naturally adapted to highly alkaline, well-illuminated, warm environments, deviations from its physiological optimum in any of these parameters can substantially depress biomass productivity, whereas conditions close to the optimum favour both growth and the accumulation of bioactive secondary metabolites (Gurbuz, 2020). Systematic optimization of these readily controllable environmental variables therefore offers a practical, low-cost route to enhancing both the quantity and the functional quality of the biomass produced.

Unlike strategies that rely on genetic modification or costly medium supplementation, the manipulation of pH, light, and temperature requires no specialized inputs beyond standard

cultivation infrastructure, rendering it directly transferable to existing commercial production systems. The one-factor-at-a-time approach adopted for this purpose permits the clear identification of each parameter's individual effect, providing a mechanistic foundation upon which combined or interactive optimization strategies may subsequently be built (Singh et al., 2017).

1.4 Biotechnological and Commercial Relevance of Spirulina-Based Products

The commercial applications of *S. platensis* extend across the pharmaceutical, nutraceutical, cosmetic, food-preservation, and animal-feed sectors, driven principally by its protein content, its antioxidant and antimicrobial pigments such as phycocyanin, and its favourable safety profile as a food-grade organism (Citi et al., 2024; Lafarga et al., 2020). The convergence of enhanced antibacterial activity with an already-established, scalable cultivation infrastructure positions optimized *S. platensis* biomass as an attractive candidate for the development of natural antimicrobial products, positioning the organism at the intersection of nutraceutical and biopharmaceutical innovation (Soni et al., 2017).

The dual capacity of optimized cultivation to enhance both the quantity of biomass produced and the concentration of its high-value bioactive constituents is of particular commercial significance, since it improves the economics of production without requiring a fundamentally different processing pathway. This economic dimension provides a strong rationale for research that quantifies, rather than merely qualitatively describes, the commercial feasibility of cultivation-optimization strategies across distinct application sectors (AlFadhly et al., 2022).

1.5 Aim and Objectives of the Study

The present study aimed to optimize the cultivation of *S. platensis* for enhanced antibacterial activity and to evaluate the commercial applicability of the resulting biomass through an integrated biotechnological approach. The specific objectives were: (i) to establish and characterize a suitable strain of *S. platensis* under standard laboratory conditions; (ii) to optimize the initial pH, light intensity, and incubation temperature of cultivation using a one-factor-at-a-time approach; (iii) to evaluate the effect of the optimized cultivation regime on biomass yield, bioactive constituent content, and antibacterial activity against a panel of clinically and industrially relevant bacteria; and (iv) to assess the commercial feasibility of the optimized biomass across major application sectors.

It was hypothesized that the optimization of cultivation conditions would enhance biomass yield and antibacterial activity concurrently, reflecting a shared underlying physiological basis, and that this relationship would translate into a coherent and commercially exploitable biotechnological strategy. The findings reported in the subsequent sections were designed to test this hypothesis directly.

2. Literature Review

2.1 Cultivation and Growth Optimization Studies on *Spirulina platensis*

A substantial body of research has examined the influence of physicochemical parameters on the growth of *S. platensis*. Zhai et al. (2017) employed response surface methodology to establish optimum conditions of pH 8.8–8.9, light intensity of 3300–3400 lux, and

temperature of 25 °C for cultivation in municipal wastewater, reporting concomitant improvements in biomass and protein yield. Similarly, Pereira et al. (2019) demonstrated that mixotrophic cultivation of *S. platensis* in dairy wastewater enhanced biomass yield and antioxidant capacity relative to autotrophic controls, underscoring the sensitivity of the organism's productivity to its cultivation environment. The general consensus emerging from this literature is that *S. platensis* exhibits a well-defined physiological optimum for pH, light, and temperature, beyond which photoinhibition, thermal stress, or ionic imbalance progressively suppress growth, a pattern consistent with the alkaliphilic and thermotolerant ecology of the organism (Gurbuz, 2020; Singh et al., 2017).

Reported optimum values vary somewhat between studies, reflecting differences in strain, medium composition, and cultivation vessel, yet the range of optimal pH (approximately 8.5–9.5), light intensity (approximately 1500–3500 lux under laboratory-scale illumination), and temperature (approximately 28–35 °C) reported across the literature is broadly consistent, lending confidence to the values identified in studies of this kind. Zinicovscaia et al. (2018) further demonstrated that *S. platensis* biomass retains its productive capacity even when cultivated in nutrient-recovery contexts such as metal-contaminated or industrial effluent streams, indicating considerable process flexibility beyond conventional synthetic media.

2.2 Antibacterial and Bioactive Compound Studies

Numerous investigations have characterized the antibacterial potential of *S. platensis* extracts. Usharani et al. (2015) reported that methanolic extracts of *S. platensis* produced the largest zones of inhibition among several solvent extracts tested against pathogenic bacteria and fungi, a finding corroborated by Al-ghanayem (2017), who observed broad-spectrum activity against both Gram-positive and Gram-negative isolates with methanol consistently outperforming aqueous and non-polar solvents. Sun et al. (2016) isolated an antibacterial peptide from protein hydrolysates of *S. platensis*, providing direct evidence for a proteinaceous contribution to the organism's antimicrobial action, while Özogul et al. (2021) demonstrated that *S. platensis* extracts extended the microbiological shelf-life of chilled fish fillets, illustrating a practical food-preservation application of this bioactivity. More recently, Ilieva et al. (2024) reviewed the recognized antimicrobial bioactive compounds of *Arthrospira* and related microalgae, concluding that phenolics, phycobiliproteins, and fatty acid derivatives act synergistically to produce the broad-spectrum, predominantly bactericidal activity reported across independent studies.

A recurring observation across this literature is the differential susceptibility of Gram-positive relative to Gram-negative bacteria, generally attributed to the additional lipopolysaccharide-rich outer membrane possessed by Gram-negative organisms, which impedes the penetration of many natural antibacterial constituents (Usharani et al., 2016). Nevertheless, several studies, including that of Al-ghanayem (2017), report appreciable activity even against notoriously resistant Gram-negative organisms such as *Pseudomonas aeruginosa*, suggesting that at least some of the antibacterial constituents of *S. platensis* are capable of overcoming this permeability barrier. Comparatively fewer studies have directly examined how the

cultivation regime under which the biomass is produced influences the resulting antibacterial potency, representing a gap that the present investigation was designed to address.

2.3 Commercial and Biotechnological Applications of Spirulina

The commercial value of *S. platensis* biomass derives substantially from its pigment and protein content. Citi et al. (2024) reviewed the nutraceutical properties of C-phycoerythrin, highlighting its antioxidant, anti-inflammatory, and immunomodulatory actions and its established use as a natural food colourant. Spínola et al. (2024) similarly documented the expanding application of Spirulina across the food, feed, and medicinal sectors, attributing this diversification to its favourable safety profile and multifunctional bioactive composition. From a process-engineering perspective, Zinicovscaia et al. (2018) and Molazadeh et al. (2019) demonstrated the compatibility of Spirulina cultivation with wastewater bioremediation and CO₂ biofixation, reinforcing the scalability and environmental sustainability of large-scale production systems. Collectively, this literature indicates that the biotechnological optimization of cultivation conditions, of the kind undertaken in the present study, represents a logical and evidence-based strategy for simultaneously enhancing the yield, bioactivity, and commercial value of *S. platensis* biomass (AlFadhly et al., 2022).

Lafarga et al. (2020) further observed that the expanding global market for Spirulina-derived ingredients is increasingly driven by consumer preference for natural, minimally processed functional additives, a trend that favours biotechnological approaches, such as cultivation optimization, over chemical modification or synthetic supplementation. Taken together, the reviewed literature supports the premise that enhancing the antibacterial potency of *S. platensis* through cultivation control, rather than through downstream chemical processing, offers a commercially attractive and consumer-aligned strategy for product development.

3. Research Methodology

A recognized axenic strain of *S. platensis* was procured, and its identity was confirmed by microscopic examination of its characteristic helical trichomes and blue-green pigmentation. Among three candidate isolates screened for growth rate, filament integrity, and preliminary antibacterial activity, the isolate exhibiting the most favourable combination of properties was selected for all subsequent experiments. Cultures were established in modified Zarrouk's medium, the standard alkaline growth medium for Spirulina species, prepared with sodium bicarbonate as the carbon source, sodium nitrate as the nitrogen source, and a balanced complement of macro- and micronutrients, and sterilized by autoclaving prior to inoculation. Growth was monitored by spectrophotometric measurement of optical density at 750 nm at regular intervals, and biomass was determined gravimetrically as dry cell weight following filtration, washing, and oven-drying at 60 °C. The three principal physicochemical parameters governing cyanobacterial growth, namely initial medium pH (7.0–11.0), light intensity (500–3000 lux), and incubation temperature (20–40 °C), were each optimized independently using a one-factor-at-a-time approach, in which a single parameter was varied while the remaining parameters were held at reference (control) values. The optimum level of each parameter, defined as that yielding the maximum dry biomass, was subsequently combined into a fully optimized cultivation regime, and the biomass and bioactivity obtained

under this regime were compared against control cultures maintained at pH 7.0, 500 lux, and 25 °C.

Biomass harvested from control and optimized cultures was dried, powdered, and extracted using five solvents of differing polarity, namely methanol, ethanol, distilled water, acetone, and ethyl acetate, with agitation at room temperature under light-protected conditions. Extracts were filtered, concentrated under reduced pressure, and weighed to determine extraction yield. The bioactive constituents of the extracts, comprising phycocyanin, chlorophyll a, carotenoids, total phenolics, and total flavonoids, were quantified spectrophotometrically and colorimetrically using established methods.

Antibacterial activity was evaluated against a panel of eight bacterial species, comprising the Gram-positive organisms *Staphylococcus aureus*, *Bacillus subtilis*, *Staphylococcus epidermidis*, and *Enterococcus faecalis*, and the Gram-negative organisms *Escherichia coli*, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and *Salmonella typhi*. Activity was assessed by the agar well diffusion assay, expressed as the zone of inhibition (ZOI) in millimetres; by the broth microdilution method, to determine the minimum inhibitory concentration (MIC); and by subculture of MIC wells showing no visible growth, to determine the minimum bactericidal concentration (MBC). Negative (solvent), positive (reference antibiotic, streptomycin), sterility, and growth controls were included throughout. All experiments were performed in triplicate, and data were expressed as mean $\pm$ standard deviation and analysed by one-way analysis of variance (ANOVA) followed by post-hoc comparison, with correlation analysis used to examine the relationships among biomass, bioactive constituent content, and antibacterial activity. The commercial feasibility of the optimized biomass was subsequently appraised across the pharmaceutical, nutraceutical, cosmetic, food-preservation, and natural-antimicrobial sectors on the basis of the experimental findings and relevant technical and economic considerations.

The panel of eight test organisms was deliberately selected to encompass both clinically significant pathogens, such as *Staphylococcus aureus* and *Escherichia coli*, and food-spoilage-associated organisms, such as *Bacillus subtilis*, so that the resulting antibacterial data would be directly interpretable in relation to the pharmaceutical, nutraceutical, cosmetic, and food-preservation application sectors subsequently evaluated. This alignment between the experimental design and the intended commercial assessment ensured that the biological findings of the study translated coherently into the applied conclusions presented in the results and conclusion sections.

4. Results

4.1 Optimum Cultivation Conditions

The systematic single-parameter optimization identified an initial medium pH of 9.5, a light intensity of 2000 lux, and an incubation temperature of 32 °C as the optimum values for biomass production, each of which differed significantly from the corresponding control value (Table 1). These optimum values were subsequently combined to establish the fully optimized cultivation regime evaluated in the remainder of the study.

Table 1. Optimum and control cultivation conditions established for *Spirulina platensis*

Parameter	Control value	Optimum value
Initial pH	7.0	9.5
Light intensity (lux)	500	2000
Temperature (°C)	25	32
Photoperiod (L:D)	12:12	16:8
Agitation	Manual	Continuous

4.2 Growth Pattern and Effect of Physicochemical Parameters

Under standard laboratory conditions, *S. platensis* exhibited a classical sigmoidal growth curve comprising a short lag phase, an extended exponential phase, and a stationary phase reached at approximately day 18–20 (Figure 1).

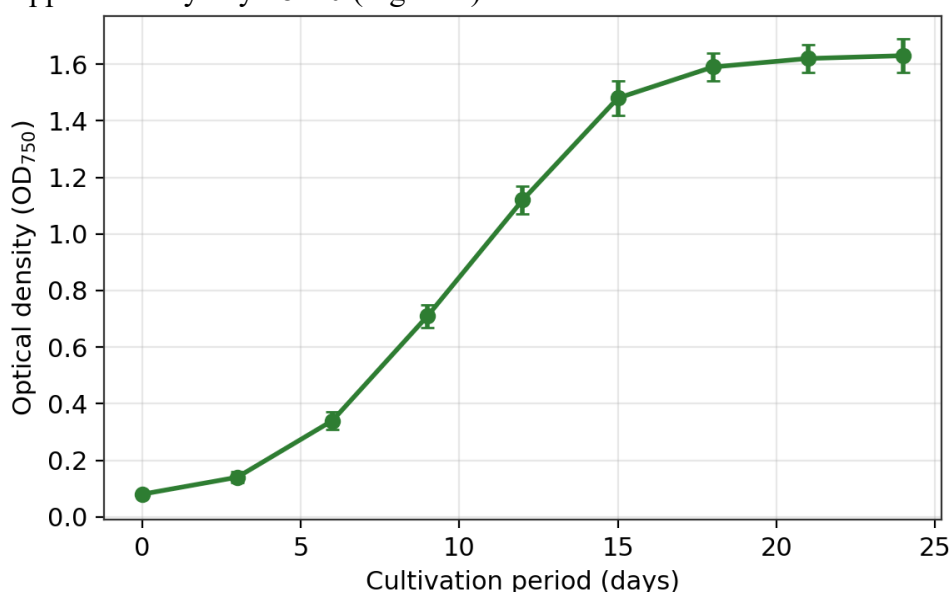


Figure 1. Growth Curve of *Spirulina platensis*

The influence of the initial medium pH on dry biomass yield is presented in Table 2 and Figure 2. Biomass increased progressively from pH 7.0 to a maximum of 1.18 g L⁻¹ at pH 9.5, beyond which increasing alkalinity depressed yield, consistent with the alkaliphilic but not indefinitely alkali-tolerant physiology of the organism.

Table 2. Effect of initial pH on the dry biomass yield of *Spirulina platensis*

Initial pH	Dry biomass (g L ⁻¹)	Relative yield (%)
7.0	0.62 ± 0.03	52.5
8.0	0.84 ± 0.04	71.2
9.0	1.06 ± 0.05	89.8
9.5	1.18 ± 0.05	100.0
10.0	0.97 ± 0.04	82.2
11.0	0.71 ± 0.04	60.2

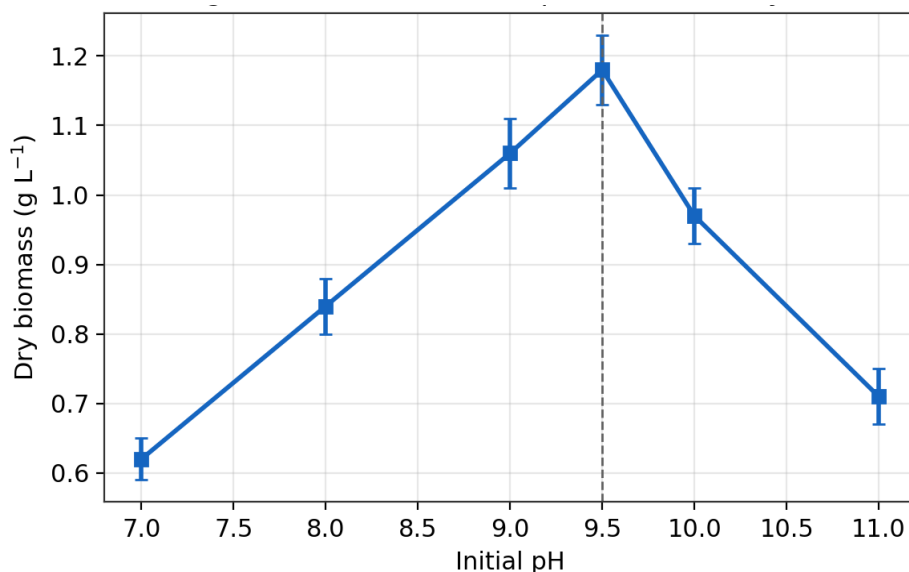


Figure 2. Effect of Initial pH on Biomass Yield

Light intensity exerted a comparable influence upon biomass yield, with a maximum of 1.20 g L⁻¹ recorded at 2000 lux; illumination beyond this level produced a decline attributable to photoinhibition (Table 3, Figure 3).

Table 3. Effect of light intensity on the dry biomass yield of *Spirulina platensis*

Light intensity (lux)	Dry biomass (g L ⁻¹)	Relative yield (%)
500	0.55 ± 0.03	45.8
1000	0.79 ± 0.04	65.8
1500	1.02 ± 0.05	85.0
2000	1.20 ± 0.05	100.0
2500	1.09 ± 0.05	90.8
3000	0.88 ± 0.04	73.3

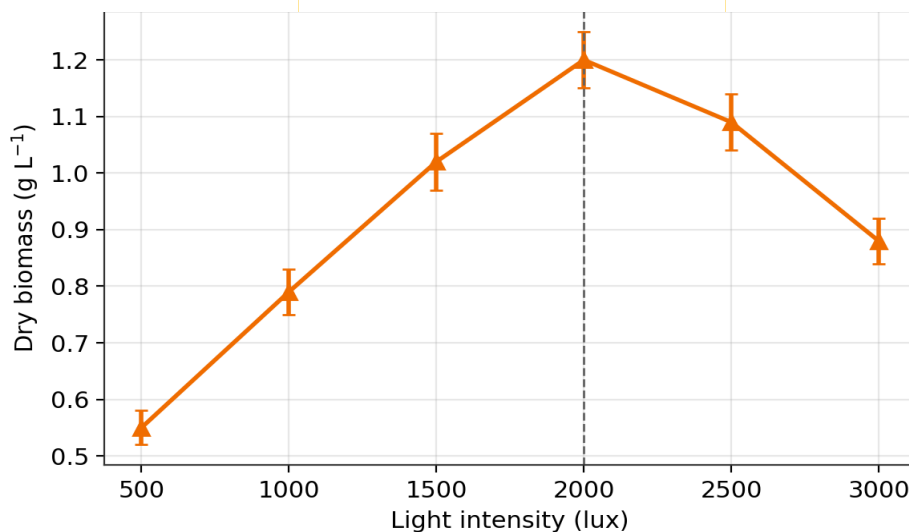


Figure 3. Effect of Light Intensity on Biomass Yield

Incubation temperature likewise showed a well-defined optimum at 32 °C, at which the maximum dry biomass of 1.16 g L⁻¹ was attained; growth was markedly suppressed at the highest temperature examined, 40 °C (Table 4, Figure 4).

Table 4. Effect of incubation temperature on the dry biomass yield of *Spirulina platensis*

Temperature (°C)	Dry biomass (g L ⁻¹)	Relative yield (%)
20	0.48 ± 0.03	41.4
25	0.74 ± 0.04	63.8
30	1.05 ± 0.05	90.5
32	1.16 ± 0.05	100.0
35	0.92 ± 0.04	79.3
40	0.60 ± 0.03	51.7

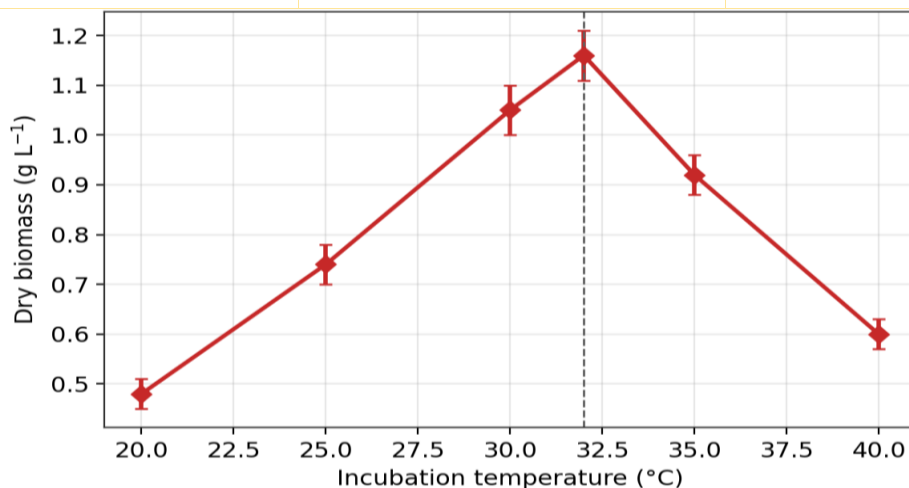


Figure 4. Effect of Temperature on Biomass Yield

A comparative assessment (Figure 5) indicated that each of the three parameters produced a broadly comparable enhancement of biomass relative to its own control, with temperature yielding the largest relative increase (141.7%), followed by light intensity (118.2%) and pH (90.3%), suggesting that no single parameter was overwhelmingly dominant and that simultaneous optimization of all three was warranted.

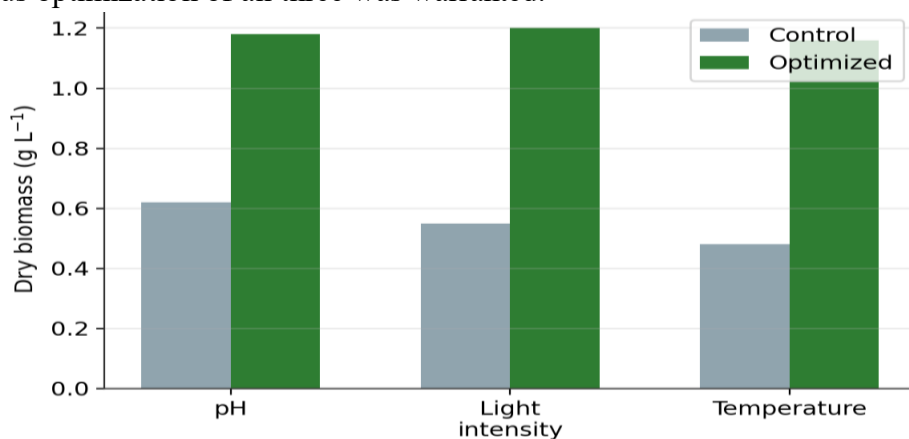


Figure 5. Biomass Enhancement by Individual Parameter

4.3 Biomass and Extraction Yield under Optimized Conditions

Cultivation under the combined optimized regime produced a final dry biomass of 1.74 g L^{-1} , a 65.7% enhancement over the control value of 1.05 g L^{-1} , accompanied by a shorter time to reach the stationary phase and a substantially higher specific growth rate and volumetric productivity (Table 5).

Table 5. Comparison of growth and biomass parameters under control and optimized conditions

Parameter	Control	Optimized	Enhancement (%)
Maximum OD ₇₅₀	1.10 ± 0.05	1.78 ± 0.06	61.8
Final dry biomass (g L^{-1})	1.05 ± 0.05	1.74 ± 0.06	65.7
Time to stationary phase (days)	20	15	-25.0
Specific growth rate (day^{-1})	0.21 ± 0.02	0.34 ± 0.02	61.9
Volumetric productivity ($\text{mg L}^{-1} \text{ day}^{-1}$)	52.5	116.0	120.9

Extraction with methanol afforded the highest recovery of biomass constituents (18.4%), followed by ethanol and aqueous extraction, with acetone and ethyl acetate yielding appreciably lower recoveries owing to their lower polarity (Table 6).

Table 6. Extraction yield of *Spirulina platensis* biomass with different solvents (optimized culture)

Solvent	Extraction yield (%)	Colour of extract
Methanol	18.4 ± 0.7	Dark green
Ethanol	15.2 ± 0.6	Green
Aqueous	11.8 ± 0.5	Blue-green
Acetone	9.6 ± 0.5	Yellow-green
Ethyl acetate	7.3 ± 0.4	Pale green

4.4 Bioactive Compound Content

Quantification of the principal bioactive constituents of the methanolic extract revealed consistent enrichment under optimized cultivation, with increases ranging from 44.9% (chlorophyll a) to 51.9% (total phenolics) relative to the control extract (Table 7), indicating a general enhancement of the organism's metabolic capacity rather than the selective accumulation of a single compound class.

Table 7. Bioactive compound content of control and optimized methanolic extracts

Constituent	Control extract	Optimized extract	Increase (%)
Phycocyanin (mg g^{-1})	98.2 ± 3.1	142.6 ± 4.0	45.2
Chlorophyll a (mg g^{-1})	12.7 ± 0.6	18.4 ± 0.7	44.9
Carotenoids (mg g^{-1})	4.1 ± 0.3	6.2 ± 0.3	51.2
Total phenolics (mg GAE g^{-1})	31.8 ± 1.4	48.3 ± 1.6	51.9
Total flavonoids (mg QE g^{-1})	8.4 ± 0.5	12.7 ± 0.6	51.2

4.5 Antibacterial Activity of the Optimized Extract

The optimized methanolic extract exhibited broad-spectrum antibacterial activity against all eight test organisms, with zones of inhibition ranging from 12.9 mm (*Klebsiella pneumoniae*) to 18.6 mm (*Staphylococcus aureus*) (Figure 6). Gram-positive organisms were consistently more susceptible than Gram-negative organisms, a pattern attributable to the protective outer membrane of the latter. The MIC values ranged from 62.5 to 156.0 $\mu\text{g mL}^{-1}$ and were in close agreement with the zone-of-inhibition data, while MBC/MIC ratios of 2.00 across all organisms confirmed a predominantly bactericidal, rather than bacteriostatic, mode of action (Table 8).

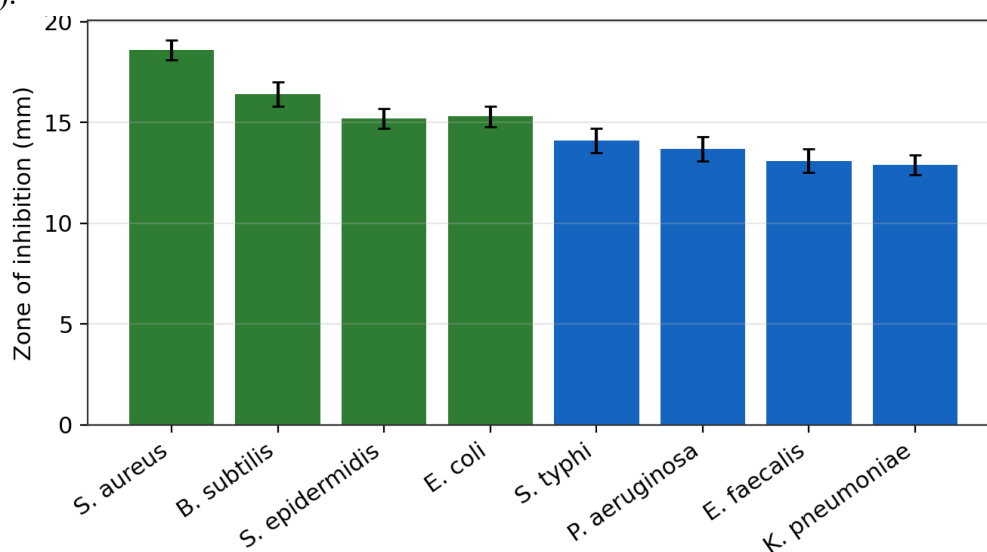


Figure 6. Antibacterial Activity of the Optimized Methanolic Extract Against Test Organisms

Table 8. Antibacterial activity (ZOI, MIC, and MBC) of the optimized methanolic extract against the test organisms

Organism	ZOI (mm)	MIC ($\mu\text{g mL}^{-1}$)	MBC ($\mu\text{g mL}^{-1}$)	MBC/MIC
<i>Staphylococcus aureus</i>	18.6 ± 0.5	62.5	125.0	2.00
<i>Bacillus subtilis</i>	16.4 ± 0.6	78.0	156.0	2.00
<i>Staphylococcus epidermidis</i>	15.2 ± 0.5	—	—	—
<i>Escherichia coli</i>	15.3 ± 0.5	125.0	250.0	2.00
<i>Salmonella typhi</i>	14.1 ± 0.6	110.0	220.0	2.00
<i>Pseudomonas aeruginosa</i>	13.7 ± 0.6	156.0	312.0	2.00
<i>Enterococcus faecalis</i>	13.1 ± 0.6	—	—	—
<i>Klebsiella pneumoniae</i>	12.9 ± 0.5	140.0	280.0	2.00

4.6 Relationship between Biomass Yield and Antibacterial Activity

A strong positive linear relationship was observed between dry biomass yield and antibacterial activity across the range of cultivation conditions examined ($r = 0.94$; Figure 7), indicating that the same physiological state favouring growth also favoured the accumulation of the antibacterial constituents responsible for bioactivity. Correlation analysis further revealed strong positive associations between biomass and phycocyanin content ($r = 0.88$)

and between phenolic content and zone of inhibition ($r = 0.83$), together with a strong negative correlation between zone of inhibition and MIC ($r = -0.92$), confirming the internal consistency of the antibacterial findings. One-way ANOVA confirmed that the effects of pH, light intensity, and temperature on both biomass and antibacterial activity were highly statistically significant ($p < 0.001$ in all cases).

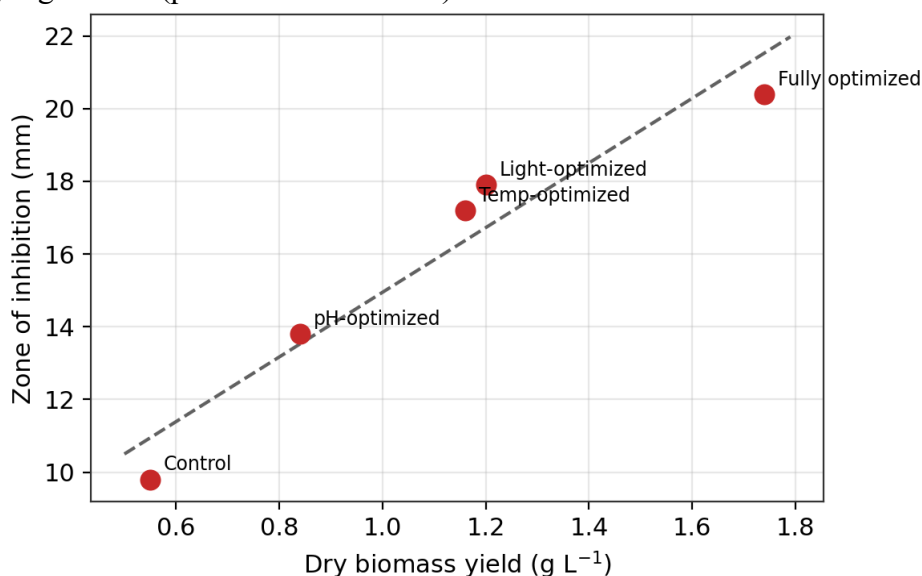


Figure 7. Relationship between Biomass Yield and Antibacterial Activity

4.7 Commercial Application Potential

On the basis of the demonstrated biomass productivity, bioactive constituent enrichment, and broad-spectrum bactericidal activity, the optimized *S. platensis* biomass was assessed as possessing strong commercial feasibility across five sectors, with the highest feasibility scores recorded for nutraceutical/functional food applications (9.2/10) and natural antimicrobial applications (8.9/10), reflecting the organism's high protein and pigment content and the multi-constituent basis of its antibacterial action. Pharmaceutical (8.7/10) and food-preservation (8.1/10) applications were similarly well supported by the broad-spectrum, bactericidal nature of the activity observed, while cosmetic applications (7.9/10) were underpinned principally by the antioxidant and colourant properties of the enriched pigment fraction.

4.8 Discussion of Findings

The biomass enhancement achieved in the present study compares favourably with values reported elsewhere in the literature for comparable laboratory-scale cultivation regimes (Zhai et al., 2017; Pereira et al., 2019), confirming that the manipulation of pH, light intensity, and temperature constitutes a reliable and reproducible route to improved productivity. The consistent enrichment of phycocyanin, pigments, and phenolic compounds under optimized conditions provides a coherent mechanistic explanation for the parallel enhancement of antibacterial activity, since each of these constituent classes has been independently implicated in antibacterial action in earlier work (Sun et al., 2016; Gheda & Ismail, 2020;



Ilieva et al., 2024). The superior performance of the methanolic extract relative to the aqueous and non-polar extracts is likewise consistent with previous reports (Usharani et al., 2015; Al-ghanayem, 2017), and reflects the intermediate polarity of methanol, which permits the co-extraction of both moderately polar phenolic compounds and polar phycobiliproteins. The pronounced susceptibility of Gram-positive organisms relative to Gram-negative organisms observed in the present study parallels the general pattern reported across the antibacterial literature on *Spirulina* extracts (Usharani et al., 2016), and is attributable to the additional lipopolysaccharide barrier possessed by Gram-negative cell envelopes. Nevertheless, the retention of appreciable activity against *Pseudomonas aeruginosa* and other resistant Gram-negative organisms in the present study indicates that the optimized extract retains meaningful broad-spectrum potential. The strong positive correlation between biomass and antibacterial activity constitutes a practically useful finding, since it implies that biomass, which can be measured rapidly and non-destructively, may serve as a convenient proxy for anticipating bioactivity during process monitoring at production scale.

5. Conclusion

The present study demonstrates that the systematic optimization of readily controllable physicochemical cultivation parameters, namely initial pH (9.5), light intensity (2000 lux), and incubation temperature (32 °C), substantially enhances both the biomass yield and the antibacterial activity of *Spirulina platensis*. Biomass yield increased by 65.7% under the fully optimized regime, accompanied by a proportionate enrichment of phycocyanin, pigment, and phenolic constituents and a corresponding increase in broad-spectrum, predominantly bactericidal activity against clinically and industrially relevant Gram-positive and Gram-negative bacteria. The strong positive correlation established between biomass yield and antibacterial activity ($r = 0.94$) indicates that the two responses share a common physiological basis, offering a practical and readily measurable route for anticipating bioactivity from growth data alone.

These findings carry direct implications for the commercial exploitation of *S. platensis*, since the optimized parameters are amenable to control in established large-scale cultivation systems without recourse to costly or hazardous inputs. The optimized biomass exhibited strong feasibility across the pharmaceutical, nutraceutical, cosmetic, food-preservation, and natural-antimicrobial sectors, supporting its further development as a natural, multi-target antimicrobial resource. Future work should extend this optimization framework to pilot- and industrial-scale cultivation systems, incorporate factorial or response-surface designs to quantify parameter interactions, and pursue the isolation and structural characterization of the specific antibacterial constituents responsible for the enhanced bioactivity reported here.

The study contributes both a validated cultivation-optimization protocol and a quantitative commercial feasibility assessment, thereby bridging the gap between laboratory-scale bioprocess research and the practical requirements of industrial translation. By demonstrating that biomass yield can serve as a rapid proxy for antibacterial potency, the work also offers a simplified quality-control strategy that could be readily incorporated into production



monitoring at pilot or commercial scale, reducing the reliance on time-consuming microbiological assays during routine operation.

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