



Response of Vegetative Growth of Three Bread Wheat Cultivars to Spraying With Spirulina Algae Extract

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ABSTRACT

Background: Seaweed is considered an environmentally friendly bio-stimulant that maintains sustainable agricultural systems and mitigates the effects of environmental pollution caused by chemical inputs. To evaluate the effectiveness of seaweed extract on the physiological growth characteristics of wheat, a field experiment was conducted during the 2023-2024 season at the College of Agricultural Engineering Sciences, University of Baghdad.

Methods: A field experiment was conducted randomized complete block design (RCBD) with three replications. The treatments included four concentrations of marine algae (Spirulina) extract (0, 2500, 5000 and 7500 mg L⁻¹) applied to three wheat cultivars (Adina, Buhouth-22 and Al-Hussein).

Result: Experimental results revealed that concentration of 5000 mg L⁻¹ was most effective, in increasing plant height, crop dry weight, crop growth rate, SPAD chlorophyll content, spike length and flag leaf area compared to other concentrations. Among the varieties, Al-Hussein cultivar demonstrated significant superiority, recording the highest relative increases across all studied parameters compared to Adina and Buhouth-22. The results indicate that the aqueous extract of seaweed acts as an effective, eco-friendly bio-stimulant for wheat, with the 5000 mg L⁻¹ concentration yielding the highest physiological performance, representing 29% of the total.

Key words: Bio-stimulants, Spirulina extract, Sustainable agriculture, Wheat varieties.

INTRODUCTION

Global food security is based on the wheat crop worldwide (Yitayew *et al.*, 2023) as it provides one-fifth of the total intake of calories and protein. Wheat is a staple food because it is rich in essential nutrients such as carbohydrates, which constitute the largest proportion, as well as proteins, vitamins and other minerals (Shewry and Hey, 2015). Wheat grains also contain chemical compounds similar to those found in some vegetables and fruits, which contributes to a balanced diet (Tiware *et al.*, 2023). However, in order to sustain agricultural systems and preserve the global environment and agriculture from pollution Recently, intense researches focused on natural aqueous extracts due to their safety and diversity of use in agriculture, such as using them as a safe alternative to pesticides (Agbo *et al.*, 2015, Diabaté *et al.*, 2014 and Jaoko *et al.*, 2020; Kamson *et al.*, 2021) or as plant growth and fitness stimulators (Al-Khafaji and Al-jubouri, 2022, Ruiz and Sanjuan, 2022) Spirulina algae considered a plant biostimulant which contains (60-70%) proteins (dry weight) (Arahou *et al.*, 2021 and Ramirez-Rodrigues *et al.*, 2021) and has a high content of amino acids and essential acids (Marrez *et al.*, 2014). A little amount of fat (5-8%) makes up its dry weight, but it contains unsaturated fatty acids most notably linolenic, linoleic, stearidonic, eicosapentaenoic, docosahexaenoic and arachidonic acids in quantities of up to 30% (Grosshagauer *et al.*, 2020; Habib *et al.*, 2008; Marrez *et al.*, 2014). Spirulina is a good source of several vitamins and minerals, including riboflavin, nicotinamide, pyridoxine, B6, folic acid, B12, cyanocobalamin, vitamin E and ascorbic acid (Jung *et al.*, 2019). It contains many

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plant hormones including 3 indole acetic acid and cytokinin, in addition to gibberellins, abscisic acid and jasmonic acid, which are important for plant growth (Amin *et al.*, 2009). Algal-based bio-stimulants are a rich source of bioactive molecules including phytohormones, minerals and vitamins that directly stimulate plant metabolism (Parmar *et al.*, 2023). These substances encourage the synthesis of chlorophyll and carotenoids, which are critical for capturing light energy (Michalak and Chojnacka, 2017 and Matinha-Cardoso *et al.*, 2023). Therefore, we notice over the past three decades, research has increasingly shifted toward the utilization of sustainable and eco-friendly compounds, such as vitamins, amino acids, seaweeds and organic substances, as viable alternatives in agricultural practices (Raju *et al.*, 2024 and Baqir *et al.*, 2024). As for varieties, the success of cultivating any crop depends primarily on sound management practices, superior field techniques and availability of suitable growth

requirements, particularly ideal environmental factors and their adaptability to local conditions. The observed variations among cultivars regarding yield and its components can be attributed to their distinct genetic architecture and their differential response to environmental conditions (Yusuf *et al.*, 2019). Thus, varieties differ in growth, productivity and response to environmental conditions according to their genetic sensitivity. Also, that varieties differ in their response to growth factors and productivity based on their genetic origin (Al-Hassan and Mahmood, 2019 and Al-Hasany *et al.*, 2019). This study seeks to address the following research questions:

1. To identify the superior cultivar that exhibits the maximum physiological growth performance among the tested wheat varieties.
2. To determine the optimal concentration of Spirulina algae extract required to achieve the highest physiological growth parameters.
3. To evaluate the synergistic effects (interaction) between Spirulina extract concentrations and wheat cultivars to identify the combination that yields the most favorable growth outcomes.
4. To quantify the percentage contribution of both algal concentrations and wheat cultivars to the overall variation in physiological growth characteristics.

MATERIALS AND METHODS

In the winter of 2023-2024, the experiment was conducted in the field of Field Crops Department at the College of Agricultural Engineering Sciences, University of Baghdad (Al-Jadriya), located at latitude 33 degrees north and longitude 44 degrees east. There were 3 replications of the RCBD (randomized complete block design). 4 different concentrations of *Spirulina* extract were the primary component (0, 2500, 5000, 7500) milligrams per liter, which were sprayed in 2 stages, first in the double node stage on the main stem and the second at flowering stage (Zadoks *et al.*, 1974). The secondary factor comprised three wheat cultivars (Adina, Buhouth-22 and Al-Hussein). Following the completion of primary and secondary tillage operations including plowing, leveling and smoothing the experimental area was partitioned into plots and irrigation channels were established. Each experimental unit measured 2 × 2 m, containing ten 2-meter-long rows with an inter-row spacing of 20 cm and a sowing depth of 5 cm. The seeding rate was maintained at 120 kg ha⁻¹. Fertilization was managed according to standard recommendations (Ali *et al.*, 2014); nitrogen was applied as urea in four split doses, while P₂O₅ was incorporated before planting at a rate of 100 kg ha⁻¹. To evaluate physiological growth parameters, random samples were collected from a protected one-meter-long interior area within each plot. Measured parameters included plant height (cm), number of branches per m², plant dry weight (g), crop growth rate (g), chlorophyll content (SPAD), spike length (cm) and flag leaf area (cm²).

The flag leaf area was determined based on the formula proposed by Thomas (1975):

$$\text{Flag leaf area} = \text{Length of the flag leaf} \times \text{Width at the center} \times 0.95$$

Statistical analysis

Using the statistical program GenStat, we compared the mathematical means using the L.S.D. approach at 5% probability level to see if there was a statistically significant difference. A split-plot analysis of variance was performed using a randomized block design, as per (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION

Physiological growth criteria

Plant height (cm)

Statistical analysis (Table 1) revealed significant effects for both spirulina extract concentrations and wheat cultivars, as well as their interaction. The 5000 mg L⁻¹ concentration achieved the highest mean plant height of 92.57 cm, significantly outperforming the other treatments with percentage increases of 14.5%, 7.9% and 5.1%, respectively. Among the cultivars, Al-Hussein exhibited a distinct superiority, recording the maximum average height of 96.60 cm, which surpassed the other varieties by 17.8% and 14.32%. Regarding the interaction, the combination of the Al-Hussein cultivar with the 5000 mg L⁻¹ concentration yielded the highest interaction mean at 102.07 cm. The pronounced effect of the 5000 mg L⁻¹ concentration on plant height can be attributed to the rich biochemical profile of Spirulina extract. It serves as a potent source of phytohormones, specifically auxins (IAA), cytokinins and gibberellins, which are essential for stimulating cell division and internodal elongation, thereby enhancing overall plant height and vegetative density (Amin *et al.*, 2009). This enhancement is further supported by the strong positive correlation coefficients (Table 2) between plant height and other physiological growth parameters, including the number of branches per m² (0.667), plant dry weight (0.773), crop growth rate (0.860), chlorophyll content (0.870), spike length (0.710) and flag leaf area (0.824).

Total number of tillers m⁻²

The statistical analysis of variance (Table 1) revealed significant effects of both spirulina extract concentrations and wheat cultivars on the number of tillers per square meter; however, their interaction was not statistically significant. The high concentration (7500 mg L⁻¹) recorded the maximum mean of 501.7 tillers m⁻², representing significant increases of 32.57% and 15.81% compared to the control and the 2500 mg L⁻¹ treatment, respectively. Notably, no significant difference was observed between the 7500 and 5000 mg L⁻¹ concentrations. Regarding the cultivars, Al-Hussein exhibited genetic superiority, producing the highest average of 470.6 tillers m⁻² and

outperforming the other varieties by 13.73% and 7.84%, respectively. Although the interaction effect was non-significant, the combination of the Al-Hussein cultivar with the 5000 mg L⁻¹ concentration yielded the highest nominal value of 537.3 tillers m⁻². The variation in tiller production can be attributed to the differential response of cultivars to growth stimulants and environmental factors based on their distinct genetic backgrounds, a finding that aligns with Al-Hasany *et al.* (2019) and Baqir and Zeboon (2026).

Dry weight at flowering (100%) g

As illustrated in Table 1, significant differences were observed across all studied factors and their interactions. The 5000 mg L⁻¹ concentration demonstrated a marked superiority, yielding the highest mean dry weight of 1628.1 g m⁻². This treatment resulted in significant percentage increases of 17.02%, 5.94% and 3.50% compared to the 0, 2500 and 7500 mg L⁻¹ concentrations, respectively. Among the cultivars, Al-Hussein recorded the maximum average dry weight of 1578.0 g m⁻², significantly outperforming the other varieties. The interaction analysis revealed that the

combination of the Al-Hussein cultivar with the 5000 mg L⁻¹ concentration produced the highest dry weight at 1690.3 g m⁻², representing a 19.0% increase over the lowest interaction mean. The observed superiority in dry matter accumulation can be attributed to enhanced energy production and metabolic efficiency, which leads to higher crop growth rates (CGR) and increased fresh matter accumulation, ultimately manifesting as higher dry weight. These findings are consistent with those reported by Mollo *et al.* (2025). The enhancement in dry matter accumulation can be collectively attributed to the increase in leaf area, tiller density and plant height. as increased plant area resulted in improved radiant energy interception and utilization, resulting in more photosynthesis and eventually more dry matter accumulation. These findings are consistent with those reported by Singh *et al.* (2024). Furthermore, this trend is corroborated by the strong positive correlation coefficients (Table 2) between dry weight and other physiological parameters, including plant height (0.773), tiller number m⁻² (0.898), CGR (1.000), chlorophyll content (0.965), spike length (0.897) and flag leaf area (0.945).

Table 1: Effect of spirulina algae concentrations and cultivars on some growth characteristics.

Treatment	Plant height (cm)	Total number of tillers m ⁻²	Plant dry weight (g m ⁻²)	Crop growth rate (CGR) (g m ⁻² day ⁻¹)	Chlorophyll content (SPAD)	Spike length (cm)	Flag leaf area (cm ²)
Spirulina algae extract (mg L⁻¹)							
0	79.28	338.3	1391.3	10.31	36.74	9.86	27.74
2500	85.20	433.2	1536.8	11.38	43.12	11.30	35.10
5000	92.57	487.9	1628.1	12.06	48.81	12.25	40.05
7500	87.93	501.7	1572.9	11.65	46.28	11.83	36.86
LSD (P=0.05)	1.13	20.49	11.90	0.08	0.50	0.18	0.59
Cultivars							
Adina	79.38	413.8	1485.3	11.00	39.95	10.33	32.72
Buhooth-22	82.76	436.4	1533.6	11.36	43.52	11.15	35.37
Al-Hussen	96.60	470.6	1578.0	11.69	47.73	12.46	36.73
LSD (P=0.05)	1.10	21.97	7.51	0.05	0.30	0.11	0.45
Adina							
0	70.88	318.7	1355.7	10.04	34.48	9.10	22.65
2500	79.18	402.6	1500.2	11.11	38.98	10.37	33.78
5000	84.35	485.7	1573.5	11.66	44.19	10.98	39.33
7500	83.11	448.2	1511.6	11.20	42.16	10.88	35.11
Buhooth22							
0	74.39	335.8	1388.4	10.28	36.25	10.18	30.34
2500	81.14	412.3	1532.8	11.35	43.27	11.15	35.22
5000	91.28	477.8	1620.5	12.00	48.12	11.82	38.63
7500	84.22	519.7	1592.6	11.80	46.45	11.45	37.27
Al-Hussen							
0	92.58	360.5	1429.8	10.59	39.50	10.31	30.23
2500	95.28	484.6	1577.3	11.68	47.10	12.39	36.30
5000	102.08	500.1	1690.3	12.52	54.11	13.96	42.19
7500	96.45	537.3	1614.6	11.96	50.23	13.16	38.20
LSD (P=0.05)	2.00	NS	15.76	0.11	0.65	0.24	0.87

Crop growth rate ($\text{g m}^{-2} \text{day}^{-1}$)

Statistical analysis (Table 1) indicated significant differences across all studied factors and their interactions. The 5000 mg L^{-1} concentration achieved the maximum mean crop growth rate (CGR) of $12.06 \text{ g m}^{-2} \text{day}^{-1}$, representing a 9.1% increase compared to the lowest concentration. Regarding the cultivars, Al-Hussein exhibited the highest average CGR at $11.69 \text{ g m}^{-2} \text{day}^{-1}$, significantly outperforming the Adina and Buhouth-22 varieties, which recorded 11.00 and $11.36 \text{ g m}^{-2} \text{day}^{-1}$ a decrease of 5.9 % and 2.8% respectively, relative to Al-Hussein. The interaction analysis revealed that the combination of the Al-Hussein cultivar with the 5000 mg L^{-1} concentration yielded the highest mean value of $12.52 \text{ g m}^{-2} \text{day}^{-1}$. This superiority can be attributed to the fact that algal-based bio-stimulants are a rich source of bioactive molecules, including phytohormones, minerals and vitamins, which directly stimulate plant metabolism. These substances promote the synthesis of chlorophyll and carotenoids, which are essential for maximizing light energy capture. These findings align with the results reported by Michalak and Chojnacka (2017) and Parmar *et al.* (2023).

Furthermore, the variation among cultivars in terms of growth, productivity and environmental responsiveness is largely governed by their distinct genetic backgrounds, as supported by Al-Hasany *et al.* (2019). These results are further corroborated by the strong positive correlation coefficients (Table 2) between CGR and other growth parameters, including plant height (0.773), tiller number m^{-2} (0.898), dry weight (1.000), chlorophyll content (0.965), spike length (0.897) and flag leaf area (0.945).

Chlorophyll content: SPAD

Statistical analysis (Table 1) revealed significant variations among the studied factors and their interactions. The 5000 mg L^{-1} concentration recorded the maximum chlorophyll content with a mean SPAD value of 48.81. This treatment significantly outperformed the other concentrations, with percentage increases of 19.55%, 11.6% and 5.2%, respectively. Among the cultivars, Al-Hussein demonstrated superior performance, achieving the highest average SPAD value of 47.73, which surpassed the other varieties by

16.3% and 8.82%. Regarding the interaction effects, the combination of the Al-Hussein cultivar with the 5000 mg L^{-1} concentration yielded the highest interaction mean of 54.11 SPAD units. The significant enhancement in chlorophyll content is likely attributed to the high concentration of amino acids and essential minerals within the extract, particularly Nitrogen (N) and Magnesium (Mg), which serve as fundamental precursors for chlorophyll biosynthesis. These results are further validated by the strong positive correlation coefficients (Table 2) between SPAD values and other physiological growth parameters, including plant height (0.870), tiller number m^{-2} (0.862), dry weight at full flowering (0.965), crop growth rate (0.965), spike length (0.960) and flag leaf area (0.890).

The spike length cm

Statistical results (Table 1) indicated significant differences among the experimental variables and their interaction. The 5000 mg L^{-1} concentration produced the maximum mean spike length of 12.25 cm, representing a 19.5% increase over the lowest concentration. Among the cultivars, Al-Hussein demonstrated superior performance, recording the longest average spike length at 12.46 cm, with significant increases of 17.09% and 10.5% compared to the other varieties. Regarding the interaction, the combination of the Al-Hussein cultivar with the 5000 mg L^{-1} concentration yielded the highest mean interaction length of 13.96 cm. This enhancement can be attributed to the role of algae extract as a potent stimulant for plant growth and cell elongation, which aligns with the findings of Al-Khafaji and Al-Jubouri (2022) and Ruiz and Sanjuan (2022). Furthermore, the high content of amino acids in the extract provides essential building blocks for protein synthesis, thereby improving nutrient uptake and enhancing vital plant growth indicators, as noted by Marrez *et al.* (2014). This superiority is further supported by the strong positive correlation coefficients (Table 2) between spike length and other physiological parameters, including plant height (0.870), tiller number m^{-2} (0.796), dry weight at full flowering (0.897), crop growth rate (0.897), chlorophyll content (0.835) and flag leaf area (0.835).

Table 2: Correlation values for the physiological characteristics of wheat growth.

Traits	Correlations						
	Plant height (cm)	Number of tillers m^{-2}	Dry weight at flowering 100% (g m^{-2})	Crop growth rate $\text{g m}^{-2} \text{d}^{-1}$	SPAD	Spike length (cm)	Flag leaf area (cm^2)
Plant height (cm)	1						
The number of tillers m^{-2}	.667**	1					
Dry weight at flowering 100% (g m^{-2})	.773**	.898**	1				
Crop growth rate $\text{g m}^{-2} \text{d}^{-1}$.773**	.898**	1.000**	1			
SPAD	.860**	.862**	.965**	.965**	1		
Spike length (cm)	.870**	.796**	.897**	.897**	.960**	1	
Flag leaf area (cm^2)	.710**	.848**	.945**	.945**	.890**	.835**	1

**Correlation is significant at the 0.01 level (2-tailed).

Flag leaf area cm²

Statistical analysis (Table 1) revealed significant variations among the studied factors and their interactions. The 5000 mg L⁻¹ concentration demonstrated marked superiority, yielding the maximum mean flag leaf area of 40.05 cm², which significantly outperformed all other tested concentrations. Among the cultivars, Al-Hussein recorded the highest average area at 36.73 cm², representing significant increases of 11.92% and 3.70% compared to the other varieties. Regarding the interaction effects, the combination of the Al-Hussein cultivar with the 5000 mg L⁻¹ concentration produced the highest interaction mean of 42.19 cm². This superiority is further corroborated by the strong positive correlation coefficients (Table 2) observed between flag leaf area and other physiological growth parameters, including plant height (0.710), tiller number m⁻² (0.848), dry weight at full flowering (0.946), crop growth rate (0.890), chlorophyll content (0.890) and spike length (0.835).

In light of the aforementioned results, the research questions posed in the introduction can be addressed as follows:

Identification of the superior cultivar

The results confirm that the Al-Hussein cultivar exhibited the highest mean values across all physiological growth parameters (Table 1), significantly outperforming the other varieties. This superiority is attributed to the cultivar's inherent genetic potential and its favorable responsiveness to growth stimulants, aligning with findings by Al-Hassan and Mahmoud (2023) and Mahmood and Al-Hassan (2023). This is further validated by the strong positive correlations observed between this cultivar and all studied physiological traits (Table 2).

Optimal bio-stimulant concentration

Regarding the second research question, the 5000 mg L⁻¹ concentration of seaweed extract yielded the most favorable results across all physiological criteria (Table 1). Notably, increasing the concentration beyond this level (7500 mg L⁻¹) appeared to be economically and productively unviable, as it did not yield proportional benefits. These findings are consistent with Baqir *et al.* (2024) and Al-Hassan *et al.* (2024). The 5000 mg L⁻¹ treatment accounted for the highest relative contribution to growth at 29%, compared to 20%, 25% and 26% for the 0, 2500 and 7500 mg L⁻¹ concentrations, respectively.

The optimal interaction treatment

In response to the third question, the interaction between the Al-Hussein cultivar and the 5000 mg L⁻¹ seaweed extract concentration proved to be the optimal combination, achieving the maximum averages for all physiological characteristics. Proportionally, the Al-Hussein variety contributed 36% to the total performance, surpassing Buhouth-22 (33%) and Adina (31%). The effectiveness of

this specific combination is reinforced by the robust positive correlation coefficients presented in Table 2.

CONCLUSION

The application of *Spirulina platensis* extract as a foliar bio-stimulant significantly enhanced the vegetative performance of wheat across multiple physiological parameters, most notably chlorophyll content. Among the evaluated genotypes, the Al-Hussein cultivar exhibited a superior responsive capacity to the treatment. The results identify 5000 mg L⁻¹ as the optimal concentration for maximizing physiological growth indicators. These findings underscore the potential of *Spirulina*-based bio-stimulants as a sustainable agricultural strategy to improve crop vigor. Further investigation is recommended to evaluate the efficacy of this bio-stimulant under diverse environmental stress conditions to support long-term agricultural sustainability.

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Disclaimers

The views and conclusions expressed in this article are solely those of the author and do not necessarily represent the views of their affiliated institutions. The author is responsible for the accuracy and completeness of the information provided, but do not accept any liability for any direct or indirect losses resulting from the use of this content.

Informed consent

No animals were used in this study.

Conflict of interest

The author declares that there are no conflicts of interest regarding the publication of this article. No funding or sponsorship influenced the design of the study, data collection, analysis, decision to publish, or preparation of the manuscript.

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