



RESEARCH ARTICLE

# Optimizing growth conditions and biostimulant application for enhanced growth, yield and quality in Butter bean (*Phaseolus lunatus L.*) cultivar KKL 1

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## Abstract

Butter beans (*Phaseolus lunatus L.*) are prized for their significant nutraceutical benefits, which are crucial in addressing malnutrition in developing countries. Given their nutritional advantages, global cultivation demand is increasing. This study aims to boost production and improve crop quality through effective agronomic practices. To minimize dependence on synthetic chemicals, the study investigates the use of biostimulants as an alternative, offering a promising method for enhancing crop growth and performance. The experiment took place under 2 different conditions: polyhouse (G<sub>1</sub>) and open field condition (G<sub>2</sub>) at Horticultural and Forestry Research Station, Kodaikanal, from December 2023 to June 2024. Various biostimulants were applied in foliar parts in different doses: control (B<sub>1</sub>) seaweed extract at 3 % (B<sub>2</sub>) and 5 % (B<sub>3</sub>), panchagavya at 3 % (B<sub>4</sub>) and 5 % (B<sub>5</sub>), vermiwash at 5 % (B<sub>6</sub>) and 10 % (B<sub>7</sub>) and fulvic acid at 6 g/L (B<sub>8</sub>) and 9 g/L (B<sub>9</sub>), each replicated 3 times. Among the treatment combinations, G<sub>1</sub>B<sub>3</sub> (seaweed extract 5 % under polyhouse condition) was most effective in all growth, yield and quality parameters including days to first flower emergence (48.50 days), number of pods per plant (96.33), pod yield (4.98 t/ha), highest soluble protein (37.36 %) than G<sub>2</sub>B<sub>1</sub> (control group under open field condition). It resulted in a greater number of days for flower emergence (57.85 days), lowest values in number of pods per plant (69.91), pod yield (2.96 t/ha) and soluble protein (24.69 %). These findings suggest that seaweed extract has great potential as a sustainable agricultural input, enhancing productivity and crop performance in butter bean cultivation. The main goals were to investigate how different biostimulants and growing conditions affect the growth and yield of butter beans and to identify effective organic growth promoters that could improve the yield and quality of butter bean crops in various growing environments. The results of this study are discussed in the following sections.

## Keywords

Biostimulant; butter bean; fulvic acid; panchagavya; seaweed extract; yield characters

## Introduction

Butter beans (*Phaseolus lunatus* L.) are leguminous crops rich in both protein and nutrients, belonging to the well-known plant family Leguminaceae. They are believed to have originated from Guatemala or nearby regions. It has adapted well to tropical environments and thrives in warm temperate climates as well as humid, sub-humid and semi-arid tropical climates. *Phaseolus lunatus*, commonly known as butter bean, lima bean or double bean, is cultivated in various regions of South Asia, notably in Maharashtra (Pune), Karnataka (Chikkamangaluru) and Tamil Nadu (Kodaikanal and Nilgiris) in India. Known for their nitrogen-fixing abilities, butter beans also contribute to soil enrichment through substantial leaf shedding, which aids in restoring soil fertility (1). Butter beans come in 2 varieties: bush and pole types. Bush varieties bear more quickly than pole lima bean varieties.

They reach a height of around 2 feet and typically have smaller seeds. Pole lima beans can reach heights of 10 to 12 feet and have large seeds; otherwise, it is called lima beans. Butter beans are highly esteemed for their nutritional profile, boasting high protein content, low sugar levels and a rich array of essential minerals. These attributes make them valuable in combating protein malnutrition, particularly in developing countries (2). They are also a significant source of vitamins and minerals. For instance, per 100 g of seed weight, butter beans typically contain 20.88 - 45 g of carbohydrates, 7.8 - 20.6 g of protein, 43 mg of starch and 1.99 - 2.9 mg of sugars. Amino acid content includes 3330 mg of glutamic acid, 1210 mg of alanine, 1280 mg of glycine and 1470 mg of lysine. In terms of minerals, they provide macro-minerals such as 111 - 366 mg of phosphorus, 508 - 1750 mg of potassium and 43 - 216 mg of magnesium. Micro-minerals found in butter beans include 2.39 - 6.8 mg of iron, 0.84 - 1.3 mg of copper and 0.95 - 1.3 mg of zinc (3, 4).

Biostimulants are potent sources of both macro and micronutrients essential for optimal plant growth, containing significant concentrations of natural plant growth regulators such as GA<sub>3</sub>, cytokinin and NAA (5). Integrating these growth-promoting biostimulants with chemical fertilizers has been shown to enhance crop productivity, reduce production costs and improve soil fertility. Liquid seaweed extract, increasingly popular globally, enriches soil with organic compounds like amino acids, trace minerals and plant growth regulators like auxin, cytokinin and gibberellins, promoting robust plant growth (6). In organic farming, panchagavya, a blend of five cow products, boosts plant immunity, mitigates common diseases and contains growth-regulating substances such as cytokinin, GA<sub>3</sub> and IAA (7). Vermiwash, derived from earthworm secretions and excretions, is valued for its micronutrient content and organic matter, demonstrating effectiveness as a foliar spray (8). Fulvic acid acts as a plant hormone and aids in buffering soil acidity and salinity, thereby enhancing plant resistance to stresses like drought and heavy metals (9). Humic acid similarly protects plants, such as maize, from water stress (10). These biostimulants collectively play a vital role in influencing the growth, yield and quality of butter beans.

## Materials and Methods

The present study detailed the effect of growing conditions and biostimulants on the growth, yield and quality of butter bean (*Phaseolus lunatus* L.) cultivar KKL 1. A field trial was carried out in both a polyhouse and an open field condition at the Horticultural and Forestry Research Station (H & FRS) in Kodaikanal (Fig. 1). The experiment took place in a polyhouse from December to April (2023-2024) and in an open field from February to June (2024). The experiment included 2 main factors: the growing conditions and the types of biostimulants used. Factor I consisted of 2 different growing conditions: polyhouse (G<sub>1</sub>) and open field (G<sub>2</sub>). Factor II involved 4 different biostimulants at varying concentrations: seaweed extract at 3 % (B<sub>2</sub>) and 5 % (B<sub>3</sub>), panchagavya at 3 % (B<sub>4</sub>) and 5 % (B<sub>5</sub>), vermiwash at 5 % (B<sub>6</sub>) and 10 % (B<sub>7</sub>) and fulvic acid at 6g/L (B<sub>8</sub>) and 9 g/L (B<sub>9</sub>). The soil in the experimental field was characterized as peaty and lateritic. This location is geographically positioned at 10°24' N latitude and 77°48' E longitude, with an elevation of 2225 m above sea level. During the study period, the average minimum temperature was 2.5 °C and the maximum was 30 °C, while the relative humidity ranged from 40 % to 100 %. The soil in the experimental field is loamy silt with a pH of approximately 5.85. Seed material used was KKL 1 butter bean high-yielding variety released during 1991; this variety was developed by Horticultural and Forestry Research Station (H & FRS) in Kodaikanal. Tender annuals are grown for their large flat, crescent, oval-shaped seeds with pod lengths of 11- 12 cm. Observations were recorded on days taken to first flowering, days taken to 50 % flowering, number of branches, plant height, number of pods per cluster, number of pods per plant, pod length (cm), pod girth (cm), 100 fresh seed weight (g), 100 dry seed weight (g), pod yield per hectare (t/ha), fresh seed yield per hectare (t/ha), dry seed yield per hectare (t/ha), soluble protein content (%), carbohydrate content (%) at the time of vegetative, flowering and yield and collected data were statistically analysed.



Fig. 1. Poly house condition (G<sub>1</sub>) and open field condition (G<sub>2</sub>).

## Results and Discussion

### Growth parameters

#### Plant height in cm at 45, 60 and 90 days after planting (DAP)

The data presented in the Table 1 shown that plant height of butter beans was significantly influenced by different biostimulants treatments which is grown under 2 different conditions. Among the 2 growing conditions the maximum plant height was recorded under polyhouse conditions ( $G_1$ ) after 45, 60, 90 DAP and final plant height (95.11 cm, 133.01 cm, 156.58 cm and 217.91 cm respectively), while the minimum plant height (89.78 cm, 116.81 cm, 148.83 cm and 196.61 cm at 45, 60, 90 DAP and final plant height respectively) was recorded under open field conditions ( $G_2$ ). Based on the biostimulant impact, highest mean plant height (100.61 cm, 136.29 cm, 168.85 cm and 229.97 cm) was observed in  $B_3$  (Seaweed extract 5 %) at 45, 60, 90 DAP and final plant height respectively. However the shortest mean plant height (85.07 cm, 112.39 cm, 136.96 cm and 185.14 cm) was observed in the control group ( $B_1$ ) at 45, 60, 90 DAP and final plant height respectively. In terms of different treatment combinations, butter beans sprayed with 5 % seaweed extract under polyhouse conditions ( $G_1B_3$ ) had the highest plant height of 104.86 cm, 147.87 cm, 176.50 cm and 254.82 cm at 45, 60, 90 DAP and final plant height respectively, whereas those treated with the control and grown in open field conditions ( $G_2B_1$ ) had the lowest height of 84.10 cm, 107.60 cm, 136.60 cm and 182.85 cm at 45, 60, 90 DAP and final plant height respectively.

The present results are contradictory to the findings (11), 0.4 % extract of *Padina minor* was the best combination to significantly enhance plant height, number of leaves and fresh weight of soybean. A similar result

reported showed that a 0.4 % extract of *Ecklonia maxima* significantly increased the vegetative growth of *Phaseolus vulgaris* (common bean) (12). One of the earlier study found that applying amino acids and seaweed extract to celeriac plants significantly increased plant height and leaf weight compared to a control group (13). This enhanced growth potential may be due to the macro- and micronutrients and other growth-promoting substances in the seaweed extract. Similarly, another study observed that foliar sprays of liquid seaweed fertilizer improved the growth and nutritional quality of okra (14). They reported significant improvements in growth parameters, including height, fresh weight and dry weight, in plants treated with these fertilizers. Additionally, various other studies (15, 16) noted increased plant size and mass with seaweed application, suggesting cytokinins as an active component.

#### Days taken to first flowering

The data in Table 2 indicate that the application of biostimulant at various concentrations significantly affected the number of days to first flowering in 2 different growing environments. Among the 2 growing conditions, the earliest flower initiation (52.12 days) was reported in plants grown under the polyhouse condition ( $G_1$ ). Meanwhile, open growing condition ( $G_2$ ) reported the maximum number of days for first flowering (55.69 days). Regarding biostimulant applications, plants sprayed with 5 % seaweed extract ( $B_3$ ) significantly produced new floral initiation at the earliest possible time (51.33 days) and control ( $B_1$ ) took the maximum floral initiation time (57.48 days). Also proven is significant variation among the treatment combinations. Specifically, plants sprayed with 5 % seaweed extract under polyhouse conditions ( $G_1B_3$ ) show a shorter duration (48.50 days) for first flower

**Table 1.** Effect of biostimulant and growing condition on plant height (cm) at 45, 60, 90 DAP and final plant height.

Treatment	Plant height (cm)											
	45 days			60 days			90 days			Final		
	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean
$B_1$	86.03 ± 1.31	84.10 ± 2.43	85.07	117.18 ± 1.08	107.60 ± 2.14	112.39	137.32 ± 4.58	136.60 ± 2.21	136.96	187.44 ± 5.00	182.85 ± 2.80	185.14
$B_2$	94.25 ± 2.55	88.20 ± 2.43	91.23	134.40 ± 3.03	116.46 ± 0.51	125.43	149.87 ± 0.13	144.31 ± 2.45	147.09	203.77 ± 2.57	191.11 ± 1.72	197.44
$B_3$	104.86 ± 4.53	96.36 ± 2.61	100.61	147.87 ± 0.13	124.71 ± 3.71	136.29	176.50 ± 0.95	161.20 ± 1.45	168.85	254.82 ± 0.46	205.14 ± 4.46	229.97
$B_4$	99.84 ± 4.23	92.29 ± 4.07	96.07	141.63 ± 4.47	122.74 ± 0.33	132.18	168.54 ± 0.91	157.71 ± 0.86	163.12	238.81 ± 4.31	201.58 ± 4.36	220.19
$B_5$	95.36 ± 2.84	89.81 ± 2.75	92.59	129.87 ± 4.22	115.27 ± 0.52	122.57	159.87 ± 5.91	145.28 ± 6.02	152.57	213.22 ± 9.03	196.34 ± 2.12	204.78
$B_6$	96.15 ± 1.99	88.26 ± 2.70	92.21	129.68 ± 1.41	114.09 ± 1.85	121.88	151.38 ± 6.14	144.26 ± 2.34	147.82	210.29 ± 3.98	192.15 ± 7.97	201.21
$B_7$	98.23 ± 0.26	91.86 ± 0.75	95.05	138.55 ± 3.75	121.54 ± 2.30	130.04	162.21 ± 2.49	153.43 ± 5.25	157.82	227.45 ± 7.38	211.10 ± 3.23	219.27
$B_8$	95.86 ± 0.09	88.80 ± 1.12	92.33	129.68 ± 3.74	115.06 ± 4.98	122.37	150.54 ± 4.62	149.88 ± 2.97	150.21	217.34 ± 3.53	193.98 ± 8.22	205.66
$B_9$	95.11 ± 1.54	88.37 ± 2.63	91.74	128.24 ± 2.31	113.85 ± 3.49	121.04	152.98 ± 1.51	146.80 ± 4.50	149.89	208.05 ± 7.13	195.28 ± 7.39	201.66
<b>Mean</b>	96.19	89.78	92.98	133.01	116.81	124.91	156.58	148.83	152.70	217.91	196.61	207.26
<b>Factor</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>
<b>SED</b>	0.81	1.73	2.45	0.773	1.72	2.43	1.12	2.39	3.221	1.44	3.05	4.32
<b>CD at 5 %</b>	1.66**	3.52**	4.98**	1.65**	3.50**	4.95**	2.29**	4.86**	6.87**	2.93**	6.21**	8.79**

NS - Non significant, \*\* - Significant, CD - Critical difference, SED - Standard error difference

**Factor 1** - Growing conditions:  $G_1$  - Poly house condition  $G_2$  - Open condition.

**Factor 2** - Biostimulants,  $B_1$  - Control,  $B_2$  - Sea weed extract 3 %,  $B_3$  - Sea weed extract 5 %,  $B_4$  - Panchagavya 3 %,  $B_5$  - Panchagavya 5 %,  $B_6$  - Vermiwash 5 %,  $B_7$  - Vermiwash 10 %,  $B_8$  - Fulvic acid 6g/L,  $B_9$  - Fulvic acid 9g/L.



initiation, while the control ( $G_2B_1$ ) took longer duration under open conditions (57.85 days). The plants treated with SLF (seaweed liquid fertilizer) led to higher chlorophyll levels, which may result from the uptake of magnesium from the SLF, a key component of chlorophyll (17). This increase in chlorophyll enhances photosynthate production and acts as a carbon source for earlier flower initiation.

### Days taken to 50 % flowering

Data presented in Table 2 showed that under polyhouse conditions ( $G_1$ ), 50 % flowering occurred earlier (70.93 days) than in open field ( $G_2$ ) conditions (76.22 days). Among the different biostimulant applications, the minimum number of days taken for 50 % flowering (68.54 days) was recorded in 5 % seaweed extract ( $B_3$ ), while the control group ( $B_1$ ) took the most days (78.69 days). In terms of interactions, plants sprayed with 5 % seaweed extract under polyhouse conditions ( $G_1B_3$ ) had the shortest time to 50 % flowering (64.02 days). The longest time (79 days) was observed in the control group under open field conditions ( $G_2B_1$ ). These findings are consistent with those of earlier study (18), where the impact of seaweed extract (*Dictyota dichotoma*) was observed when applied on *Abelmoschus esculantus*. Similar results were also reported in tomatoes (19), demonstrating that higher potassium levels can significantly accelerate flower initiation. Their findings highlighted that seaweed liquid extract from *Sargassum crassifolium* is rich in potassium relative to other macronutrients and growth regulators that contribute to enhancing flower production through effective pollination, which is facilitated by potassium. These findings were corroborated with the earlier finding (20), which demonstrated that potassium enhances the rate of pollen germination and the formation of pollen tubes in *Arabidopsis* via controlling the turgor pressure within the pollen tube. Because potassium plays a more important role as a major osmotica in pollen tube growth than in

pollen germination. The growth of the pollen tube involves a rapid expansion of cytoplasmic volume, necessitating adequate potassium ions to maintain optimal cytoplasmic solute concentrations.

### Number of branches

As per Table 2, a greater number of branches per plant (4.25) were recorded under polyhouse conditions ( $G_1$ ), whereas plants grown in the open field ( $G_2$ ) had fewer branches per plant (3.64). Among the biostimulant applications, the 5 % seaweed extract ( $B_3$ ) resulted in a higher number of branches per plant (4.42), while the control group ( $B_1$ ) had the minimum number of branches (3.20). Among all treatment combinations, the highest number of branches (4.79) was observed with the 5 % seaweed extract under polyhouse conditions ( $G_1B_3$ ), and the lowest number (3.19) was noticed in the control group under open field conditions ( $G_2B_1$ ). Earlier study (21) noted that the application of seaweed extract (SWE) enhanced root growth by inducing the expression of an auxin-related gene, which elevated auxin levels and consequently promoted cell division and expansion. As a result, cambial activity is stimulated, leading to earlier interfascicular cambium development and an increase in the number of vascular bundles. These changes in internodal cells are associated with a significant increase in water uptake, osmotic pressure and the transport of sugars and nitrogenous compounds (22). These factors are effectively linked to the vegetative growth of plants, leading to an increase in the number of branches per plant. This finding is supported by an earlier study (11) where 0.4 % extract of *Padina minor* was the most effective combination, significantly increasing the number of branches and fresh weight of soybean plants.

### Yield parameters

Significant difference was noted between the various growing conditions, biostimulant application and their interactions for the yield parameters of butter beans.

**Table 2.** Effect of biostimulant and growing condition on days taken to first flowering, days taken to 50 % flowering and number of branches.

Treatment	Days taken to first flowering			Days taken to 50 % flowering			Number of branches		
	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean
$B_1$	57.11 ± 0.24	57.85 ± 2.22	57.48	78.38 ± 3.53	79.00 ± 0.31	78.69	3.21 ± 0.13	3.19 ± 0.04	3.20
$B_2$	54.56 ± 1.38	55.20 ± 1.09	54.88	73.68 ± 1.59	77.90 ± 2.18	75.79	3.90 ± 0.12	3.36 ± 0.10	3.62
$B_3$	48.50 ± 1.49	54.56 ± 1.57	51.53	64.02 ± 2.14	73.07 ± 0.59	68.54	4.79 ± 0.18	4.06 ± 0.09	4.42
$B_4$	49.87 ± 0.40	55.90 ± 1.26	52.88	68.00 ± 2.15	74.55 ± 0.47	71.27	4.68 ± 0.06	3.86 ± 0.13	4.27
$B_5$	53.02 ± 1.58	54.56 ± 0.54	53.79	71.32 ± 1.22	75.84 ± 3.35	73.58	4.21 ± 0.13	3.64 ± 0.07	3.92
$B_6$	50.52 ± 0.18	56.51 ± 2.44	53.51	70.22 ± 2.47	77.28 ± 0.98	73.74	4.47 ± 0.17	3.99 ± 0.09	4.23
$B_7$	50.80 ± 0.05	56.12 ± 1.47	53.46	69.18 ± 1.68	75.23 ± 1.36	72.20	4.53 ± 0.13	3.76 ± 0.12	4.14
$B_8$	52.01 ± 1.59	54.20 ± 0.73	53.10	70.99 ± 0.58	76.19 ± 2.20	73.59	4.37 ± 0.09	3.57 ± 0.09	3.96
$B_9$	52.73 ± 0.95	56.33 ± 1.12	54.53	72.65 ± 1.11	76.92 ± 2.22	74.78	4.13 ± 0.08	3.41 ± 0.14	3.77
<b>Mean</b>	52.12	55.69	53.90	70.93	76.22	73.57	4.25	3.64	3.95
<b>Factor</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>
<b>SED</b>	0.36	0.77	1.09	0.52	1.11	1.58	0.02	0.06	0.08
<b>CD at 5 %</b>	0.74**	1.57**	2.22**	1.07**	2.27**	3.21**	0.05**	0.12**	0.17**

NS - Non significant, \*\* - Significant, CD - Critical difference, SE.d - Standard error difference

**Factor 1** - Growing conditions:  $G_1$  - Poly house condition  $G_2$  - Open condition.

**Factor 2** - Biostimulants,  $B_1$  - Control,  $B_2$  - Sea weed extract 3 %,  $B_3$  - Sea weed extract 5 %,  $B_4$  - Panchagavya 3 %,  $B_5$  - Panchagavya 5 %,  $B_6$  - Vermiwash 5 %,  $B_7$  - Vermiwash 10 %,  $B_8$  - Fulvic acid 6g/L,  $B_9$  - Fulvic acid 9g/L.

### Number of pods per cluster

Analysis of the data in Table 3 revealed that, the highest number of pods per cluster (5.99) was observed under the polyhouse ( $G_1$ ) condition, while the lowest (5.02) was noted in the open field ( $G_2$ ). Among the biostimulants tested, seaweed extract 5 % ( $B_3$ ) led to the highest number of pods per cluster at 6.28 and the control group ( $B_1$ ) had the lowest count at 4.33. Specifically, the combination of seaweed extract 5 % under polyhouse conditions ( $G_1B_3$ ) resulted in the maximum number of pods per cluster (6.78) and the minimum number of pods per plant (4.41) was recorded in the control group under open field conditions ( $G_2B_1$ ). The findings are consistent with those of earlier findings (23, 24), where increased bean pod yield, number of pods per plant and number of pods per cluster were reported after applying foliar sprays of crude extracts from *Macrocystis integrifolia* and *Ecklonia maxima*.

### Number of pods per plant

Significant differences were noted in the number of pods per plant in butter beans treated with various biostimulants under both open and protected growing conditions (Table 3). Under polyhouse conditions ( $G_1$ ), plants produced more pods (87.85) compared to those in the open field ( $G_2$ ), which had 78.35 pods. Among the biostimulants, seaweed extract 5 % ( $B_3$ ) resulted in the highest number of pods per plant (91.10) and the control group ( $B_1$ ) had the fewest pods (71.23). In terms of interactions, the maximum number of pods per plant (96.33) was observed with seaweed extract 5 % under protected conditions ( $G_1B_3$ ) and the minimum number of pods (69.91) was recorded in the control group

under open field conditions ( $G_2B_1$ ). The present results are contradictory to the findings of another study (25) who applied liquid extracts of marine algae as a soil drench to cluster bean plants and observed significant improvements in yield-related parameters, including the number of pods per plant, pod weight and pod length. The enhancements in seaweed-treated plants are believed to be linked to the hormonal substances present in the extracts, particularly cytokinins. In vegetative plant organs, cytokinins are associated with nutrient partitioning, while in reproductive organs, high levels of cytokinins may be related to nutrient allocation.

### Pod length (cm)

The investigation on pod length in butter beans sprayed with different biostimulants and grown under open and protected conditions is summarized (Table 3). The longest pod length was significantly recorded under polyhouse conditions ( $G_1$ ) at 11.95 cm, compared to 11.07 cm in open field conditions ( $G_2$ ). Among the biostimulants, seaweed extract 5 % ( $B_3$ ) resulted in the maximum pod length of 12.18 cm, with the control group ( $B_1$ ) having the shortest pod length of 10.65 cm. In terms of interactions, seaweed extract 5 % ( $B_3$ ) produced the maximum pod length of 12.70 cm under polyhouse conditions ( $G_1B_3$ ), while the minimum pod length of 10.62 cm was recorded in the control group under open conditions ( $G_2B_1$ ) (Table 3). This finding is supported by an earlier findings (25) using liquid extracts of marine algae as a soil drench for cluster bean plants and observing significant improvements in yield parameters, including the pod length and pod weight.

**Table 3.** Effect of biostimulant and growing condition on number of pods per plant, pod length (cm), pod girth (cm), Number of pods per cluster.

Treat ment	Number of pods per cluster			Number of pods per plant			Pod length (cm)			Pod girth (cm)		
	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean
$B_1$	4.25 ± 0.05	4.41 ± 0.12	4.33	72.56 ± 2.04	69.91 ± 1.88	71.23	10.69 ± 0.41	10.62 ± 0.13	10.65	3.21 ± 0.03	3.14 ± 0.13	3.17
$B_2$	5.56 ± 0.21	4.57 ± 0.02	5.06	85.21 ± 0.15	74.31 ± 0.87	79.75	11.55 ± 0.02	10.75 ±0.18	11.14	3.77 ± 0.16	3.27 ± 0.07	3.52
$B_3$	6.78 ± 0.30	5.79 ± 0.10	6.28	96.33 ± 3.47	85.88 ± 1.78	91.10	12.70 ± 0.09	11.67 ± 0.01	12.18	4.33 ± 0.14	3.85 ± 0.09	4.09
$B_4$	6.59 ± 0.20	5.41 ± 0.24	5.99	92.16 ± 2.82	82.53 ± 0.15	87.34	12.57 ± 0.01	11.39 ± 0.03	11.97	4.24 ± 0.14	3.69 ± 0.05	3.96
$B_5$	6.01 ± 0.21	5.12 ± 0.07	5.56	87.53 ± 1.89	79.70 ± 3.02	83.61	11.83 ± 0.51	11.15 ± 0.26	11.49	3.98 ± 0.10	3.51 ± 0.01	3.74
$B_6$	6.24 ± 0.25	4.72 ± 0.05	5.48	89.90 ± 0.89	75.62 ± 1.23	82.76	12.16 ± 0.52	10.87 ± 0.38	11.51	4.09 ± 0.15	3.36 ± 0.13	3.72
$B_7$	6.38 ± 0.07	5.28 ± 0.02	5.83	90.40 ± 1.55	81.02 ± 2.34	85.71	12.44 ± 0.48	11.22 ± 0.46	11.82	4.15 ± 0.01	3.58 ± 0.07	3.86
$B_8$	6.19 ± 0.09	5.08 ± 0.02	5.63	89.01 ± 0.88	78.37 ± 1.55	83.69	11.95 ± 0.12	11.07 ± 0.06	11.51	4.01 ± 0.04	3.47 ± 0.12	3.74
$B_9$	5.94 ± 0.20	4.84 ± 0.10	5.39	86.72 ± 3.67	77.85 ± 2.88	82.28	11.74 ± 0.31	10.92 ± 0.06	11.33	3.92 ± 0.09	3.41 ± 0.04	3.66
<b>Mean</b>	5.99	5.02	5.50	87.75	78.35	83.05	11.95	11.07	11.51	3.96	3.47	3.72
<b>Factor</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>
<b>SED</b>	0.04	0.08	0.12	0.58	1.24	1.75	0.07	0.16	0.23	0.02	0.05	0.07
<b>CD at 5 %</b>	0.08**	0.18**	0.25**	1.19**	2.52**	3.57**	0.16**	0.34**	0.48**	0.05**	0.10**	0.15*

NS - Non significant, \*\* - Significant, CD - Critical difference, SE.d - Standard error difference

**Factor 1** - Growing conditions:  $G_1$  - Poly house condition  $G_2$  - Open condition.

**Factor 2** - Biostimulants,  $B_1$  - Control,  $B_2$  - Sea weed extract 3 %,  $B_3$  - Sea weed extract 5 %,  $B_4$  - Panchagavya 3 %,  $B_5$  - Panchagavya 5 %,  $B_6$  - Vermiwash 5 %,  $B_7$  - Vermiwash 10 %,  $B_8$  - Fulvic acid 6g/L,  $B_9$  - Fulvic acid 9g/L.

### Pod girth (cm)

Significant differences in pod girth of butter beans were observed when sprayed with different biostimulants under open and protected environmental conditions. According to Table 3, the maximum pod girth was recorded under protected conditions ( $G_1$ ) at 3.96 cm, while the minimum pod girth of 3.47 cm was noted under open conditions ( $G_2$ ). Among all biostimulant treatments seaweed extract 5 % ( $B_3$ ) resulted in the maximum pod girth of 4.09 cm, with the control group ( $B_1$ ) having the minimum pod girth of 3.17 cm. Among all interactions, seaweed extract 5 % under protected conditions ( $G_1B_3$ ) recorded the maximum pod girth of 4.33 cm and the minimum pod girth of 3.14 cm recorded in the control group under open field conditions ( $G_2B_1$ ). These findings are consistent with those of earlier findings (26, 27). This could be attributed to the relatively higher carbohydrate content, which may have promoted growth rates, thereby increasing the number of pods, the weight of individual pods and other pod-related traits.

### Pod yield per hectare (t/ha)

Significant differences in pod yield per hectare were observed among all butter beans grown under 2 different conditions (Table 4 and Fig. 2). The highest yield per hectare (4.27 t/ha) was achieved under protected ( $G_1$ ) conditions, whereas the lowest yield (3.85 t/ha) was recorded under open ( $G_2$ ) field conditions. Seaweed extract 5 % ( $B_3$ ) resulted in the highest yield per hectare (4.58 t/ha) and the minimum yield (3.12 t/ha) was observed in the control group ( $B_1$ ). Regarding interactions, butter beans treated with seaweed extract 5 % under polyhouse conditions ( $G_1B_3$ ) produced the maximum pod yield per hectare (4.98 t/ha) and the lowest yield (2.96 t/ha)

was noted in the control group under open field conditions ( $G_2B_1$ ). Previous studies (23, 24) have shown that foliar application of crude extracts from *Macrocystis integrifolia* and *Ecklonia maxima* enhances pod yield in beans. This finding is supported (28) where applying Kelpak 66 seaweed liquid extract foliarly increased bean yields by 24 %. This result aligns with earlier findings, where seaweed extracts were shown to significantly improve potato plant growth and yields (29) and seaweed liquid fertilizer (SLF) from *Rosenvingea intricata* was found to enhance okra yields (30). Besides, foliar applications of seaweed extract increased soybean yields, with the highest yields from seaweed-treated plants (31). Similarly, seaweed extracts have been found to increase crop yields due to their ability to boost chlorophyll levels (32).

### Fresh seed yield per hectare (t/ha)

The Table 4, indicated that there were significant variation among the nine biostimulant treatments, different growing conditions and their interactions on fresh seed yield per hectare. The polyhouse ( $G_1$ ) growing condition resulted in the highest fresh seed yield per hectare (1.61 t/ha), while the lowest yield (1.55 t/ha) was recorded under open ( $G_2$ ) environmental conditions. Butter beans treated with seaweed extract 5 % ( $B_3$ ) produced the highest fresh seed yield per hectare (1.84 t/ha) and the control group ( $B_1$ ) had the lowest yield (1.45 t/ha). Among the interactions, seaweed extract 5% under polyhouse conditions ( $G_1B_3$ ) achieved the highest yield per hectare (1.92 t/ha) and the lowest yield (1.44 t/ha) was recorded in the control group under open conditions ( $G_2B_1$ ). These results were similar to some of the earlier findings (14, 25, 27, 33).

**Table 4.** Effect of biostimulant and growing condition on pod yield per hectare (t/ha), fresh seed yield per hectare (t/ha) and dry seed yield per hectare (t/ha).

Treatment	Pod yield per hectare (t/ha)			Fresh seed yield per hectare (t/ha)			Dry seed yield per hectare (t/ha)		
	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean
$B_1$	3.27 ± 0.08	2.96 ± 0.13	3.12	1.45 ± 0.04	1.44 ± 0.02	1.45	0.64 ± 0.01	0.62 ± 0.02	0.63
$B_2$	3.99 ± 0.17	4.04 ± 0.11	4.02	1.55 ± 0.05	1.60 ± 0.07	1.58	0.76 ± 0.03	0.75 ± 0.19	0.76
$B_3$	4.98 ± 0.19	4.18 ± 0.08	4.58	1.92 ± 0.07	1.76 ± 0.04	1.84	1.28 ± 0.04	1.08 ± 0.03	1.18
$B_4$	4.73 ± 0.05	4.12 ± 0.08	4.43	1.77 ± 0.03	1.75 ± 0.06	1.76	1.19 ± 0.03	1.02 ± 0.03	1.11
$B_5$	4.20 ± 0.03	3.91 ± 0.04	4.06	1.50 ± 0.03	1.47 ± 0.01	1.49	0.84 ± 0.02	0.70 ± 0.03	0.77
$B_6$	4.33 ± 0.02	3.86 ± 0.02	4.10	1.61 ± 0.04	1.44 ± 0.01	1.53	0.98 ± 0.04	0.73 ± 0.02	0.86
$B_7$	4.68 ± 0.11	4.10 ± 0.05	4.39	1.72 ± 0.01	1.59 ± 0.01	1.66	1.06 ± 0.02	0.93 ± 0.02	0.99
$B_8$	4.47 ± 0.12	4.02 ± 0.17	4.25	1.52 ± 0.06	1.50 ± 0.05	1.51	0.92 ± 0.04	0.89 ± 0.03	0.91
$B_9$	3.81 ± 0.09	3.45 ± 0.11	3.63	1.48 ± 0.06	1.43 ± 0.04	1.46	0.69 ± 0.01	0.67 ± 0.02	0.68
Mean	4.27	3.85	4.06	1.61	1.55	1.58	0.93	0.82	0.87
Factor	G	B	G×B	G	B	G×B	G	B	G×B
SED	0.02	0.06	0.08	0.01	0.02	0.03	0.01	0.02	0.03
CD at 5 %	0.05**	0.12**	0.17**	0.02**	0.04**	0.06**	0.02**	0.05**	0.08**

NS - Non significant, \*\* - Significant, CD - Critical difference, SED - Standard error difference,

Factor 1 - Growing conditions:  $G_1$  - Poly house condition  $G_2$  - Open condition,

Factor 2 - Biostimulants,  $B_1$  - Control,  $B_2$  - Sea weed extract 3 %,  $B_3$  - Sea weed extract 5 %,  $B_4$  - Panchagavya 3 %,  $B_5$  - Panchagavya 5 %,  $B_6$  - Vermiwash 5 %,  $B_7$  - Vermiwash 10 %,  $B_8$  - Fulvic acid 6g/L,  $B_9$  - Fulvic acid 9g/L.

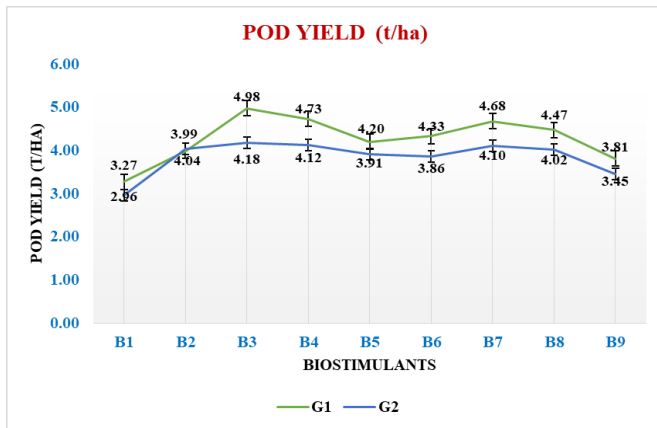


Fig. 2. Effect of biostimulant and growing condition on pod yield per ha (t/ha).

### Dry seed yield per hectare (t/ha)

Significant differences in butter bean dry seed yield per hectare were observed among different biostimulants, growing conditions and their interactions. As per the Table 4, the polyhouse ( $G_1$ ) growing condition resulted in the highest dry seed yield per hectare (0.93 t/ha), while the lowest yield (0.82 t/ha) was recorded under open ( $G_2$ ) environmental conditions. Butter beans treated with seaweed extract 5 % ( $B_3$ ) produced the highest dry seed yield per hectare (1.18 t/ha) and the control group ( $B_1$ ) had the lowest yield (0.63 t/ha). Among the interactions, butter beans treated with seaweed extract 5 % under protected conditions ( $G_1B_3$ ) achieved the highest yield per hectare (1.28 t/ha) and the lowest yield (0.62 t/ha) was recorded in the control group under open conditions ( $G_2B_1$ ). This finding is supported (30), who studied the effects of foliar applications of various concentrations of seaweed extract on soybean yield. They found that these applications significantly enhanced yield parameters, with the highest grain yield recorded at a 15 % seaweed extract concentration. This was followed by a 12.5 % concentration, resulting in 57 % and 46 % increases in yield respectively compared to the control.

## Biochemical parameters

### Soluble protein content (%)

The combined impact of biostimulants and growing conditions significantly affected the soluble protein content in the cultivars. Polyhouse conditions ( $G_1$ ) yielded the highest protein content at 29.30 %, compared to 28.90 % in open field conditions ( $G_2$ ). Butter beans treated with 5 % seaweed extract ( $B_3$ ) achieved the highest protein content of 34.26 %, while the control ( $B_1$ ) had the lowest at 29.54 %. Among the interaction effects, butter beans with 5 % seaweed extract under polyhouse conditions ( $G_1B_3$ ) showed the highest protein content at 37.36 %. Conversely, the control under open conditions ( $G_2B_1$ ) had the lowest protein content at 24.69 % (Table 5 and Fig. 3). Similar results align with various other studies (25, 34), who found that using marine liquid extracts as soil drench on cluster bean plants significantly increased biochemical parameters like photosynthetic pigments, protein content and amino acids. The rise in photosynthetic pigments may be due to magnesium, a key component for chlorophyll synthesis or the increase in the number and size of chloroplasts and better grana development.

### Carbohydrate content (%)

There was a slight but notable difference in carbohydrate content among butter beans treated with different biostimulants under 2 growing conditions and their interactions (Table 5 and Fig. 4). The highest carbohydrate content was observed under polyhouse conditions ( $G_1$ ) at 42.60 %, while the open field conditions ( $G_2$ ) recorded a slightly lower carbohydrate content at 39.58 %. Among the biostimulants, seaweed extract at 5 % ( $B_3$ ) resulted in the highest carbohydrate content at 43.64 % and the control group ( $B_1$ ) had the lowest carbohydrate content at 37.70 %. In terms of interactions, seaweed extract at 5 % under polyhouse conditions ( $G_1B_3$ ) had the highest carbohydrate content at 45.31 % and the lowest carbohydrate content

Table 5. Effect of biostimulant and growing condition on soluble protein content (%), carbohydrate content (%).

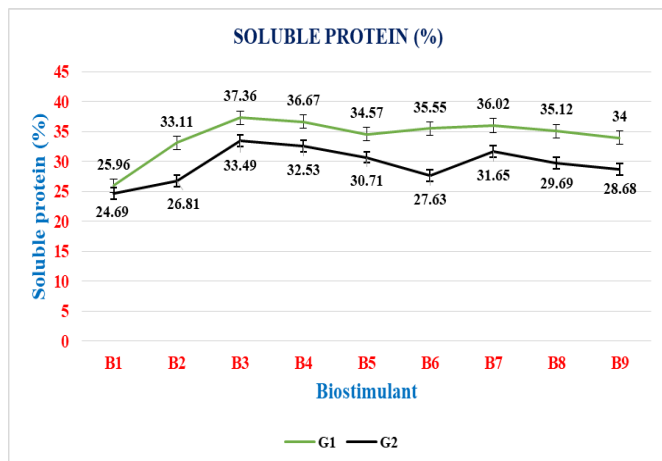
Treatment	Soluble protein content (%)			Carbohydrate content (%)		
	$G_1$	$G_2$	Mean	$G_1$	$G_2$	Mean
$B_1$	25.96 ± 0.22	24.69 ± 0.16	25.32	37.52 ± 0.67	37.90 ± 0.45	37.70
$B_2$	33.11 ± 0.24	26.81 ± 0.46	29.96	41.17 ± 0.31	38.14 ± 0.47	39.65
$B_3$	37.36 ± 1.38	33.49 ± 0.36	35.42	45.31 ± 0.82	41.99 ± 0.62	43.64
$B_4$	36.67 ± 0.20	32.53 ± 0.09	34.60	44.59 ± 0.87	40.75 ± 0.52	42.67
$B_5$	34.57 ± 1.18	30.71 ± 0.39	32.64	42.23 ± 0.38	39.80 ± 0.49	41.01
$B_6$	35.55 ± 1.09	27.63 ± 0.25	31.59	43.54 ± 0.43	38.81 ± 0.24	41.17
$B_7$	36.02 ± 0.55	31.65 ± 0.83	33.83	43.93 ± 0.27	40.22 ± 0.24	42.07
$B_8$	35.12 ± 0.82	29.69 ± 0.48	32.40	43.13 ± 0.41	39.53 ± 0.69	41.32
$B_9$	34.00 ± 1.26	28.68 ± 0.47	31.34	42.03 ± 0.39	39.09 ± 0.08	40.56
Mean	34.26	29.54	31.90	42.60	39.58	41.09
Factor	<b>G</b>	<b>B</b>	<b>G×B</b>	<b>G</b>	<b>B</b>	<b>G×B</b>
SED	0.18	0.39	0.56	0.13	0.29	0.41
CD at 5 %	0.38**	0.80**	1.14**	0.28**	0.60	0.84**

NS - Non significant, \*\* - Significant, CD - Critical difference, SED - Standard error difference,

Factor 1 - Growing conditions:  $G_1$  - Poly house condition  $G_2$  - Open condition,

Factor 2 - Biostimulants,  $B_1$  - Control,  $B_2$  - Sea weed Extract 3 %,  $B_3$  - Sea weed Extract 5 %,  $B_4$  - Panchagavya 3 %,  $B_5$  - Panchagavya 5 %,  $B_6$  - Vermiwash 5 %,  $B_7$  - Vermiwash 10 %,  $B_8$  - Fulvic acid 6g/L,  $B_9$  - Fulvic acid 9g/L.



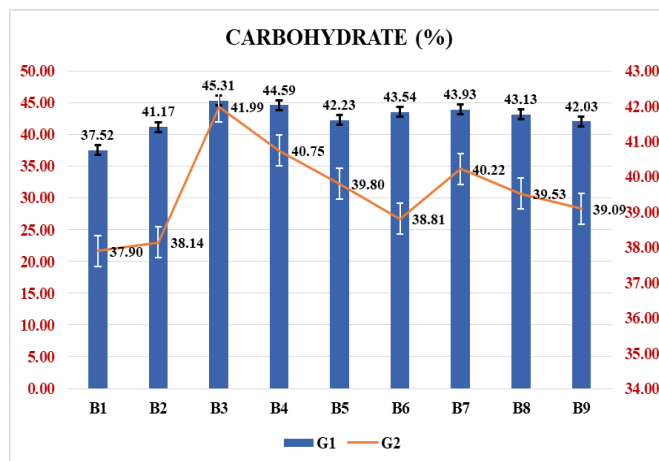


**Fig. 3.** Effect of biostimulant and growing condition on soluble protein content (%).

was recorded in the control group under open field conditions ( $G_2B_1$ ) at 37.90 %. Similar results were earlier reported (25), where liquid marine extracts as soil drenches for cluster bean plants significantly reduced sugar levels. Similarly, another study (13) observed that amino acids and seaweed extracts increased total sugar content in celeriac plants compared to controls, with the effect being more pronounced at higher application rates and no significant difference between the 2 treatments. These findings align with earlier studies (31), which reported that foliar application of liquid seaweed fertilizers (LSF) enhanced the nutritional quality of okra, increasing levels of carbohydrate, protein and dietary fiber. Similarly, one of the earlier works found that seaweed extract increased both seed yield and nutritional quality in green gram plants, enhancing carbohydrate and protein content compared to the control group (14).

## Conclusion

In the 2 different growing conditions, the polyhouse ( $G_1$ ) condition achieved the highest vegetative growth, yield and quality. The butter beans were treated with various biostimulants, including seaweed extract, panchagavya, vermiwash and fulvic acid at different concentrations. Seaweed extract at 5 % concentration showed superior performance in terms of yield and growth attributes in both polyhouse ( $G_1$ ) and open field ( $G_2$ ) conditions. The study clearly demonstrates that using seaweed extract biostimulants is highly beneficial, as they promoted early maturation and improved quality parameters such as higher crude fiber, carbohydrate levels. A biostimulant significantly boosts the growth and yield of butter beans by enhancing their resilience to environmental stresses and improving seed quality. It also decreases reliance on chemical treatments. Looking ahead, the application of biostimulants is expected to play a crucial role in sustainable agriculture by promoting more resilient crops and reducing the environmental impact of farming practices. As research advances, biostimulants may become increasingly sophisticated, offering even greater benefits for crop productivity and ecological health.



**Fig. 4.** Effect of biostimulant and growing condition on carbohydrate (%).

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## Authors' contributions

MCS: Writing of original draft and conceptualization. RKLK: Revision of draft, inclusion of tables and figures, proof reading. MKKTS: Revision, formatting and Supervision. All the authors read and approved the final version of the manuscript.

## Compliance with ethical standards

**Conflict of interest:** Authors do not have any conflict of interests to declare.

**Ethical issues:** There is no any ethical issues.

## Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work, I used CHATGPT in order to PARAPHRASING. For data running and analysis, I used AGRES software. After using this tool/service, I reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

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