

Original Research Article

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# Physiological and Nutritional Responses of French Bean to Seaweed Extract Application

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## ABSTRACT

### Keywords

Seaweed biostimulant; *Phaseolus vulgaris*; foliar application; nutrient uptake; pod yield

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The study was carried out employing initial laboratory trial to identify the optimum concentration of seaweed extract (SWE) for seed treatment followed by field experiment at ZARS, GKVK, Bengaluru during *Kharif* 2024 with ten treatments replicated thrice in RCBD to evaluate best quantity of SWE for foliar application. It was compared with absolute control, recommended dose of fertilizers (RDF) and vegetable special spray. Application of RDF + seed treatment with SWE @ 2000 ppm + foliar spray of SWE @ 2 ml L<sup>-1</sup> at 15 and 45 DAS (T<sub>8</sub>) recorded significantly higher growth (per plant branch number-12.7, leaf area-590.9 cm<sup>2</sup> and SPAD value-57.88) yield parameters (per plant pod number-28.8 and pod weight-191.54 g) and higher pod yield (27.53 t ha<sup>-1</sup>) followed by T<sub>10</sub> (RDF + foliar spray of vegetable special @ 2 g L<sup>-1</sup>). The same treatment recorded the higher uptake of primary (N: 159.8 kg ha<sup>-1</sup>, P: 58.3 kg ha<sup>-1</sup>, K: 118.1 kg ha<sup>-1</sup>). Whereas, higher micronutrient uptake was observed in T<sub>10</sub> (Fe: 459.2 g ha<sup>-1</sup>, Zn: 149.2 g ha<sup>-1</sup>, Cu: 96.4 g ha<sup>-1</sup> and Mn: 142.3 g ha<sup>-1</sup>).

## Introduction

Meeting the rising demand for nutritious food under conditions of shrinking cultivable land and increasing climate variability remains a major challenge for modern agriculture. Grain legumes and vegetable legumes play a vital role in ensuring food and nutritional security due to their high protein content, dietary fibre and essential micronutrients. Among them, French bean (*Phaseolus vulgaris* L.) is an important short-duration vegetable

legume widely cultivated in India for fresh pod consumption. Despite its economic and nutritional importance, productivity of French bean is often constrained by declining soil fertility, imbalanced fertilizer use and low nutrient use efficiency.

The excessive and continuous reliance on chemical fertilizers has resulted in soil degradation, nutrient imbalance and environmental pollution, necessitating the adoption of sustainable nutrient management strategies.

In this context, plant biostimulants have gained considerable attention as eco-friendly inputs capable of enhancing nutrient uptake efficiency, crop growth and yield without adversely affecting soil health. Seaweed extracts are among the most widely used biostimulants due to their rich composition of polysaccharides, amino acids, vitamins, micronutrients and naturally occurring plant growth regulators such as auxins, cytokinins and gibberellins (Khan *et al.*, 2009; Barsanti *et al.*, 2022).

Extracts derived from brown seaweeds, particularly *Ascophyllum nodosum*, have been reported to improve seed germination, vegetative growth, photosynthetic efficiency, nutrient uptake and tolerance to abiotic stresses in several crops (Crouch *et al.*, 1990; Jannin *et al.*, 2013). In addition, seaweed-based biostimulants contribute to improved soil structure and microbial activity, thereby supporting long-term soil fertility. The response of crops to seaweed extracts, however, largely depends on the concentration, method and timing of application. Seed treatment and foliar application are the most common and effective modes of delivery, either alone or in combination.

Considering the limited systematic information on the combined effect of seed treatment and foliar application of seaweed extract on French bean, the present study was undertaken to standardize the optimum concentration of seaweed extract for seed treatment and foliar application and to evaluate their individual and combined effects on growth, yield, nutrient uptake and economics of French bean cultivation under field conditions.

## Materials and Methods

### Experimental site and design

A field experiment was conducted during the Kharif season of 2024 at the L-Block, Integrated Farming System Unit, University of Agricultural Sciences (UAS), GKVK, Bengaluru, Karnataka, India. The experimental site is located in the Eastern Dry Zone of Karnataka. The experiment was laid out in a randomized complete block design with ten treatments and three replications. (Table 1.)

### Treatment Details

T<sub>1</sub>: Absolute control

T<sub>2</sub>: Recommended Dose of fertilizers

T<sub>3</sub>: RDF + FA - SWE 1 ml L<sup>-1</sup>

T<sub>4</sub>: RDF + FA - SWE 2 ml L<sup>-1</sup>

T<sub>5</sub>: RDF + FA - SWE 3 ml L<sup>-1</sup>

T<sub>6</sub>: RDF + ST - SWE @ 2000 ppm

T<sub>7</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 1 ml L<sup>-1</sup>

T<sub>8</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup>

T<sub>9</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L<sup>-1</sup>

T<sub>10</sub>: RDF + FA - Vegetable special @ 2 g L<sup>-1</sup>

### Note:

**RDF** - Recommended dose of fertilizer (63:100:75 NPK kg ha<sup>-1</sup>)

**SWE** - Seaweed extract

**ST** - Seed treatment **FA** - Foliar application at 15 & 45 DAS

**DAS**-Days after sowing

### Soil characteristics

Prior to the initiation of the experiment, a composite soil sample was collected from the 0–15 cm soil depth and analyzed for physico-chemical properties following standard procedures. The soil of the experimental site was red sandy loam in texture and classified under *Alfisols*. The soil reaction was slightly acidic (pH 6.5) with an electrical conductivity of 0.14 dS m<sup>-1</sup> and organic carbon content of 3.9 g kg<sup>-1</sup>. The available nutrient status of the soil was medium in nitrogen (278.73 kg ha<sup>-1</sup>), phosphorus (62.4 kg ha<sup>-1</sup>) and potassium (185.8 kg ha<sup>-1</sup>) (Table 2.)

### Seaweed extract and treatments

The seaweed extract used in the study was derived from *Ascophyllum nodosum* and characterized for its chemical composition prior to application (Table 3.). An initial laboratory study was conducted to determine the optimum concentration for seed treatment, and 2000 ppm for 30 minutes was found to be ideal. The treatments comprised seed treatment and foliar application of seaweed extract at different concentrations in combination with the recommended dose of fertilizers (63:75:100 kg N:P<sub>2</sub>O<sub>5</sub>:K<sub>2</sub>O ha<sup>-1</sup>). Foliar applications were carried out at 15 and 45 days after sowing. The treatments also included an absolute control and a commercial vegetable special spray for comparison.

### Crop management

French bean variety ‘Ayush’ was used for the experiment. Recommended agronomic practices were followed uniformly for all treatments throughout the cropping period. Observations on growth parameters

were recorded at harvest, while pod yield was computed on a hectare basis. Plant and soil samples were analyzed for nutrient content using standard analytical methods (Jackson, 1973; Piper, 1966; Subbiah and Asija, 1956; Bray and Kurtz, 1945; Lindsay and Norvell, 1978). Nutrient uptake was calculated by multiplying nutrient concentration with corresponding dry matter yield.

### Statistical analysis

The data obtained were subjected to statistical analysis following the procedure described by Gomez and Gomez (1984). Treatment means were compared using critical difference values at the 5 per cent level of significance.

## Results and Discussion

### Growth parameters

The data on growth parameters as influenced by seed treatment and foliar application of seaweed extract are presented in Table 3. Among the treatments, RDF + Seed treatment with SWE 2000 ppm + Foliar spray of 2 ml L<sup>-1</sup> MESWE at 15 & 45 DAS (T<sub>8</sub>) recorded significantly higher growth parameters such as plant height (67.3 cm), number of branches plant<sup>-1</sup> (12.7), leaf area plant<sup>-1</sup> (590.9 cm<sup>2</sup>) and SPAD value (51.47) which was followed by application of RDF + FA - Vegetable special @ 2 g L<sup>-1</sup> with 65.7 cm, 11.8, 564.8 cm<sup>2</sup> and 49.53. However, the lower growth parameters were recorded in no fertilizer application (absolute control) with the plant height (47.9 cm), number of branches plant<sup>-1</sup> (7.9), leaf area plant<sup>-1</sup> (470.4 cm<sup>2</sup>) and SPAD value (38.50) presented in Table 4. The foliar application of seaweed extract (SWE) significantly enhanced the vegetative growth of French bean, as indicated by increase in growth parameters. This overall improvement might be due to the action of auxins, gibberellins, cytokinin and essential nutrients found in seaweed extract that promoted cell division, elongation, chlorophyll synthesis and lateral bud development. Enhanced physiological activity, including better translocation of photosynthates and nitrogen assimilation, also contributed to the increased chlorophyll content reflected in higher SPAD readings. These findings are in line with Dilavarnaik *et al.*, (2017), who reported similar improvements in growth parameters with seaweed extract application in other crops. Overall, SWE acted as an effective bio stimulant, improving the vegetative vigour of French bean.

### Pod Yield (t ha<sup>-1</sup>)

The pod yield of French bean varied significantly with application of seaweed extract (*Ascophyllum nodosum*) as seed treatment and foliar application. (Table 4.). The higher pod yield of 27.53 t ha<sup>-1</sup> was recorded in treatment T<sub>8</sub> (RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup> at 15 & 45 DAS). The Treatments T<sub>10</sub> (RDF + FA - Vegetable special @ 2 g L<sup>-1</sup> at 15 and 45 DAS) and T<sub>9</sub> (RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L<sup>-1</sup>) also recorded high yields (26.13 t ha<sup>-1</sup> and 24.49 t ha<sup>-1</sup>, respectively) which were statistically comparable to T<sub>8</sub>, demonstrating no significant difference among these superior treatments. However, lower pod yield (10.13 t ha<sup>-1</sup>) was recorded in absolute control (T<sub>1</sub>) (Table 4; Figure 1).

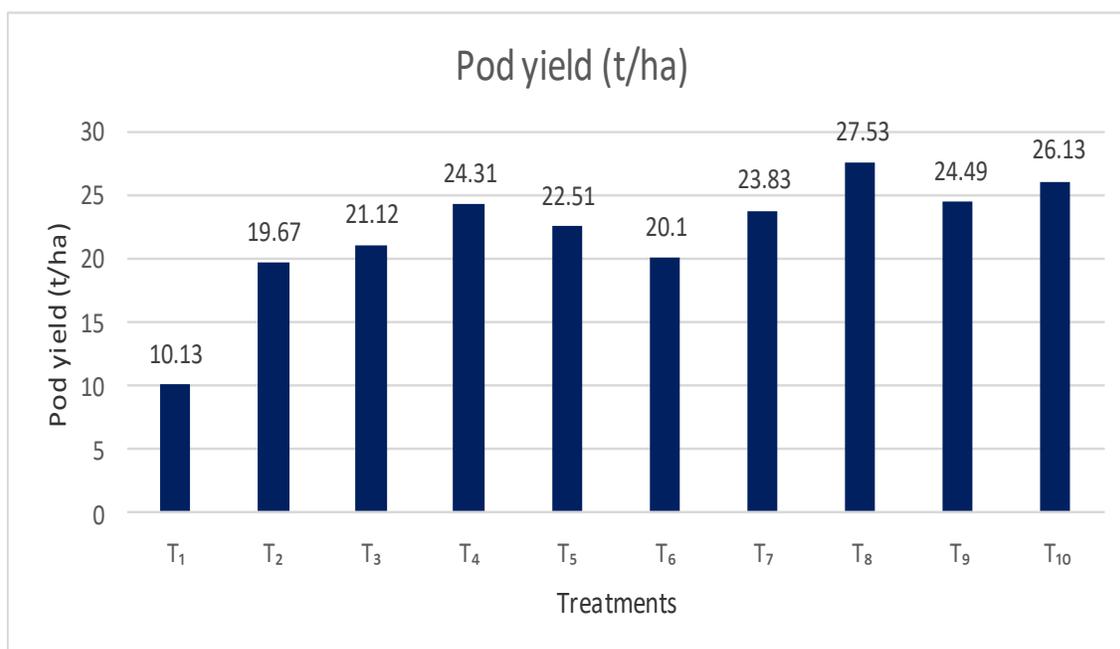
The substantial increase in pod yield with SWE might be attributed to enhanced enzymatic activity and improved physiological efficiency. Seaweed extract is known to stimulate nitrate reductase, amylase and dehydrogenase activities, which support better nitrogen assimilation, carbohydrate mobilization and energy availability during pod setting.

The presence of polysaccharides, amino acids and chelated micronutrients in the extract likely improved pollen viability, flower retention and reduced flower drop, resulting in a higher number and well-filled pods. These findings align with the observations reported by Temple and Bomke (1988), who noted an increase in bean pod yield following foliar application of crude extracts derived from *Macrocystis integrifolia* and *E. maxima*. Rathore *et al.*, (2009) investigated various concentrations of seaweed extract applied to soybean and found significant enhancement in yield-related traits. The highest grain yield was obtained with 15% seaweed extract, followed by 12.5 per cent, showing 57 per cent and 46 per cent yield increase respectively, over the control group

### Economics of seaweed extract application

The B: C ratio, which reflects the profitability of treatments, varied from 1.45 in T<sub>1</sub> to 3.82 in T<sub>8</sub>. The treatment T<sub>8</sub> (RDF + MESWE seed treatment at 2000 ppm kg<sup>-1</sup>+ Foliar spray of 2 ml L<sup>-1</sup> MESWE at 15 & 45 DAS) recorded the maximum B:C ratio (3.82), followed closely by T<sub>10</sub> (3.73), T<sub>4</sub> (3.47), T<sub>9</sub> (3.44), and T<sub>7</sub> (3.43) and lower B:C recorded in control (1.45).

**Fig.1** Effect of seaweed extract on pod yield of French bean



**Table.1** Initial soil physico chemical properties of the experimental site

Sl. No.	Particulars	Values
1	Sand	61.43
2	Silt	24.50
3	Clay	14.07
4	Textural class	Sandy loam
1	Soil pH (1:2.5)	6.5
2	EC (1:2.5) (dSm <sup>-1</sup> )	0.14
3	Organic Carbon (g kg <sup>-1</sup> )	3.9
4	Available N (kg ha <sup>-1</sup> )	278.73
5	Available P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	62.4
6	Available K <sub>2</sub> O (kg ha <sup>-1</sup> )	185.8
7	Exchangeable Ca [c mol (p <sup>+</sup> ) kg <sup>-1</sup> ]	5.2
8	Exchangeable Mg [c mol (p <sup>+</sup> ) kg <sup>-1</sup> ]	2.6
9	Available S (mg kg <sup>-1</sup> )	19.4
10	DTPA extractable Fe (mg kg <sup>-1</sup> )	23.14
11	DTPA extractable Mn (mg kg <sup>-1</sup> )	19.86
12	DTPA extractable Zn (mg kg <sup>-1</sup> )	0.89
13	DTPA extractable Cu (mg kg <sup>-1</sup> )	0.62
14	Available B (mg kg <sup>-1</sup> )	0.46

**Table.2** Chemical Properties of seaweed extract

Sl. No	Parameters	Values
1	pH	7.7
2	EC (dSm <sup>-1</sup> )	0.30
3	Nitrogen (%)	0.98
4	Phosphorus (%)	0.03
5	Potassium (%)	0.70
6	Calcium (%)	0.375
7	Magnesium (%)	0.110
8	Zinc (mg kg <sup>-1</sup> )	13.12
9	Iron (mg kg <sup>-1</sup> )	528.5
10	Manganese (mg kg <sup>-1</sup> )	7.925
11	Copper (mg kg <sup>-1</sup> )	15.6

**Table.3** Effect of seaweed extract on growth parameters of French bean at harvest

Treatments	Plant height (cm)	No. of Branches	Leaf area (cm <sup>2</sup> )	SPAD value
T <sub>1</sub> : Absolute control	47.9	7.9	470.4	38.50
T <sub>2</sub> : RDF (63:75:100 N:P:K kg ha <sup>-1</sup> )	52.5	9.3	529.7	45.80
T <sub>3</sub> : RDF + FA - SWE 1 ml L <sup>-1</sup>	53.9	10.1	542.9	45.89
T <sub>4</sub> : RDF + FA - SWE 2 ml L <sup>-1</sup>	58.1	11.1	545.2	49.23
T <sub>5</sub> : RDF + FA - SWE 3 ml L <sup>-1</sup>	54.6	11.0	536.7	46.38
T <sub>6</sub> : RDF + ST - SWE @ 2000 ppm	53.1	9.6	535.1	48.03
T <sub>7</sub> : RDF + ST - SWE @ 2000 ppm + FA - SWE 1 ml L <sup>-1</sup>	62.9	10.5	559.2	45.85
T <sub>8</sub> : RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L <sup>-1</sup>	67.3	12.7	590.9	51.47
T <sub>9</sub> : RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L <sup>-1</sup>	63.9	11.3	560.0	49.50
T <sub>10</sub> : RDF + FA - Vegetable special @ 2 g L <sup>-1</sup>	65.7	11.8	564.8	49.53
S.Em. ±	1.42	0.27	21.77	1.14
C.D. (0.05)	4.23	0.80	64.70	3.38

**Note:**

**RDF** - Recommended dose of fertilizer (63:100:75 NPK kg ha<sup>-1</sup>) **SWE** - Seaweed extract

**ST** - Seed treatment **FA** - Foliar application at 15 & 45 DAS **DAS**-Days after sowing

**Table.4** Effect of seaweed extract on pod yield and nutrient uptake of French bean at harvest

Treatments	Pod Yield (t ha <sup>-1</sup> )	Benefit cost ratio
<b>T<sub>1</sub>: Absolute control</b>	10.13	1.45
<b>T<sub>2</sub>: RDF (63:75:100 N:P:K kg ha<sup>-1</sup>)</b>	19.67	2.71
<b>T<sub>3</sub>: RDF + FA - SWE 1 ml L<sup>-1</sup></b>	21.12	2.93
<b>T<sub>4</sub>: RDF + FA - SWE 2 ml L<sup>-1</sup></b>	24.31	3.47
<b>T<sub>5</sub>: RDF + FA - SWE 3 ml L<sup>-1</sup></b>	22.51	3.08
<b>T<sub>6</sub>: RDF + ST - SWE @ 2000 ppm</b>	20.10	2.79
<b>T<sub>7</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 1 ml L<sup>-1</sup></b>	23.83	3.43
<b>T<sub>8</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup></b>	27.53	3.82
<b>T<sub>9</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L<sup>-1</sup></b>	24.49	3.44
<b>T<sub>10</sub>: RDF + FA - Vegetable special @ 2 g L<sup>-1</sup></b>	26.13	3.73
<b>S.Em. ±</b>	<b>0.57</b>	
<b>C.D. (0.05)</b>	<b>1.71</b>	

**Note:**

**RDF** - Recommended dose of fertilizer (63:100:75 NPK kg ha<sup>-1</sup>) **SWE** - Seaweed extract

**ST** - Seed treatment **FA** - Foliar application at 15 & 45 DAS **DAS**-Days after sowing

**Table.5** Effect of seaweed extract on pod yield, total primary nutrient and micronutrient uptake

Treatments	Total nutrient uptake (kg ha <sup>-1</sup> )			Total nutrient uptake (g ha <sup>-1</sup> )			
	N	P	K	Fe	Zn	Cu	Mn
<b>T<sub>1</sub>: Absolute control</b>	58.01	24.32	56.86	205.99	76.77	38.79	68.84
<b>T<sub>2</sub>: RDF (63:75:100 N:P:K kg ha<sup>-1</sup>)</b>	118.42	40.88	82.61	309.74	98.10	56.21	92.70
<b>T<sub>3</sub>: RDF + FA - SWE 1 ml L<sup>-1</sup></b>	138.35	43.79	97.40	358.32	104.96	65.14	104.47
<b>T<sub>4</sub>: RDF + FA - SWE 2 ml L<sup>-1</sup></b>	147.20	48.83	104.28	390.05	123.07	76.09	115.77
<b>T<sub>5</sub>: RDF + FA - SWE 3 ml L<sup>-1</sup></b>	140.91	47.54	100.96	371.52	113.41	68.36	107.45
<b>T<sub>6</sub>: RDF + ST - SWE @ 2000 ppm</b>	127.68	43.35	89.24	341.47	102.35	60.77	100.96
<b>T<sub>7</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 1 ml L<sup>-1</sup></b>	146.81	51.53	106.31	393.96	127.27	76.70	119.53
<b>T<sub>8</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup></b>	159.85	58.27	118.13	432.62	142.93	92.58	134.64
<b>T<sub>9</sub>: RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L<sup>-1</sup></b>	152.70	53.57	111.83	415.31	133.34	85.63	126.65
<b>T<sub>10</sub>: RDF + FA - Vegetable special @ 2 g L<sup>-1</sup></b>	155.31	54.71	113.56	459.17	149.23	96.45	142.27
<b>S.Em. ±</b>	<b>2.32</b>	<b>0.98</b>	<b>1.69</b>	<b>6.42</b>	<b>2.05</b>	<b>1.27</b>	<b>1.95</b>
<b>C.D. (0.05)</b>	<b>6.89</b>	<b>2.92</b>	<b>5.02</b>	<b>19.07</b>	<b>6.09</b>	<b>3.76</b>	<b>5.79</b>

**Note:**

**RDF** - Recommended dose of fertilizer (63:100:75 NPK kg ha<sup>-1</sup>) **SWE** - Seaweed extract

**ST** - Seed treatment **FA** - Foliar application at 15 & 45 DAS **DAS**-Days after sowing

This higher profitability may be attributed to enhanced growth, yield attributes and nutrient uptake resulting in increased marketable yield without a proportionate increase in cost of cultivation.

## Nutrient uptake

The total primary and secondary nutrient uptake of French bean varied significantly across treatments. The higher uptake of N (159.85 kg ha<sup>-1</sup>), P (58.27 kg ha<sup>-1</sup>) and K (118.13 kg ha<sup>-1</sup>) was recorded with application of RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup> at 15 & 45 DAS (T<sub>8</sub>) followed by foliar application of vegetable special @ 2 g L<sup>-1</sup> at 15 & 45 DAS (T<sub>10</sub>). While, the lower uptake occurred in the control (Table 5). The superior performance of T<sub>8</sub> could be attributed to the combined effect of SWE applied as seed priming and foliar feeding, which readily supplied available nutrients and bioactive compounds such as cytokinin, auxins, vitamins, organic acids and natural chelators. These constituents likely enhanced nutrient solubility, membrane permeability, root activity and assimilation efficiency, ultimately improving nutrient translocation to actively growing tissues.

The findings of the current study align with those of [Shah et al., \(2013\)](#), who observed that foliar application of *Kappaphycus alvarezii* and *Gracilaria edulis* extracts improved nutrient uptake in wheat, with the higher uptake of N, K, Ca, Mg and S recorded at 5% *G. edulis* sap, and maximum N and P uptake achieved at 7.5 per cent *K. alvarezii* sap concentration.

Significant differences were observed in Fe, Zn, Cu and Mn uptake in pods and haulms of French bean under SWE treatments. The higher micronutrient Fe (459.17 g ha<sup>-1</sup>), Zn (149.23 g ha<sup>-1</sup>), Cu (96.45 g ha<sup>-1</sup>) and Mn (142.27 g ha<sup>-1</sup>) was observed in T<sub>10</sub> (RDF + FA - Vegetable special @ 2 g L<sup>-1</sup> at 15 and 45 DAS), followed by T<sub>8</sub> (RDF + ST - SWE @ 2000 ppm + FA - SWE 2 ml L<sup>-1</sup> at 15 & 45 DAS) and T<sub>9</sub> (RDF + ST - SWE @ 2000 ppm + FA - SWE 3 ml L<sup>-1</sup> at 15 & 45 DAS), while the control recorded the lower values (Table 5).

The superior uptake in SWE treated plants could be attributed to enhanced foliar absorption supported by bioactive constituents such as natural chelators, cytokinin, auxins and betaines, which improved leaf permeability and nutrient mobility. Foliar application also bypasses soil fixation losses, leading to greater micronutrient availability. Additionally, SWE induced

metabolic stimulation likely enhanced enzymatic activities and photosynthetic processes, increasing the demand and assimilation of micronutrients like Fe and Zn that are essential for chlorophyll synthesis and efficient physiological functioning. The application of seaweed sap has been found to promote root growth, likely due to certain components such as organic acids that can chelate nutrients ([Crouch et al., 1990](#)), along with the upregulate of genes responsible for nitrogen uptake and assimilation ([Jannin et al., 2013](#)). In vegetable special treatment which enhances the micronutrient uptake in french bean it might be due to availability of balanced micronutrient availability. The results of the present study are in close agreement with the findings of [Basavarajeshwari Patil et al., \(2007\)](#), who reported that enhanced growth and yield were attributed to improved micronutrient availability and efficient absorption through foliar application, leading to better physiological activity and nutrient utilization.

In conclusion, the present investigation demonstrated that the integrated application of seaweed extract as seed treatment (2000 ppm) followed by foliar spray at 2 ml L<sup>-1</sup> at 15 and 45 days after sowing, in combination with the recommended dose of fertilizers, significantly enhanced growth, pod yield, nutrient uptake and economic returns of French bean.

The improved performance under this treatment can be attributed to enhanced physiological activity, better nutrient availability and improved nutrient use efficiency induced by bioactive compounds present in the seaweed extract. Although foliar application of vegetable special spray also resulted in higher yield and micronutrient uptake, the seaweed extract-based treatment proved to be more cost-effective.

Therefore, the combined use of seaweed extract as seed treatment and foliar application can be recommended as a sustainable and profitable nutrient management strategy for French bean cultivation under similar agro-climatic conditions.

## Author Contributions

Ammaisha Bagawan: Investigation, formal analysis, writing—original draft. S. Channakeshava: Validation, methodology, writing—reviewing. H. M. Atheek ur rehaman:—Formal analysis, writing—review and editing. G. G. Kadalli: Investigation, writing—reviewing.

## Data Availability

The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

## Declarations

**Ethical Approval** Not applicable.

**Consent to Participate** Not applicable.

**Consent to Publish** Not applicable.

**Conflict of Interest** The authors declare no competing interests.

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