

Standardization of Selective Biofertilizer-Based Seed Treatments for Enhanced Growth and Yield in French Bean (*Phaseolus vulgaris* L.)

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ABSTRACT: French bean (*Phaseolus vulgaris* L.) is a vital legume crop valued for its nutritional content and economic significance in global agriculture. However, poor field emergence, suboptimal crop stand, and low yield often limit its productivity. To overcome this, seeds are subjected to Pre-sowing treatment with biofertilizers, such as Phosphate Solubilizing Bacteria (PSB), *Rhizobium*, *Azospirillum*, and *Trichoderma*, which offer a sustainable approach to enhance seed germination, plant growth, and yield. This study, conducted during 2022-23 at the Field Experimentation Centre and Notified State Seed Testing Laboratory, Department of Genetics and Plant Breeding, aimed to identify optimal dose of pre-sowing seed treatment with biofertilizers for improving the agronomic performance of French bean variety Ellora 11. among all the pre sowing seed treatment, *rhizobium* 12% for 6hrs showed better performance and yield. The findings are significant for promoting sustainable agricultural practices by reducing reliance on chemical fertilizers, thereby contributing to improved crop productivity and environmental health.

Keywords: French bean, *Azospirillum*, PSB, *Rhizobium*, *Trichoderma*, Pre-sowing Seed treatment

INTRODUCTION

One of the most extensively cultivated legumes worldwide, the French bean (*Phaseolus vulgaris* L.), also known as snap beans, green beans, or string beans, is prized for its direct application in human diets [1]. It is a multipurpose crop that can be eaten while they are young and tender, shell beans when they reach physiological maturity, and dry beans when they are completely grown [2].

French beans are well-liked due to their nutritional content, short growing season, and soft, edible pods [3]. Climbing (pole) and dwarf (bush) are the two primary growth kinds; bush forms are more prevalent since they mature more quickly and don't require support [4].

French bean pods are typically plucked before the seeds are fully grown, while they are still soft and meaty. These pods are perfect for whole consumption because they have a lower fiber content, are typically stringless, and have thick, juicy walls [5]. They can be green, golden, or purple, and they may or may not have spots. French beans must first be cooked (thermally processed). It

boosts nutritional availability, decreases antinutrients, and improves digestibility in addition to taste and texture [6].

French beans are high in dietary fiber, water, proteins, vitamins, and minerals. They are excellent for low-carb diets and appropriate for diabetics because they are low in fat and sodium, cholesterol-free, and have a low glycaemic index [7].

French beans provide a number of vital elements in addition to amino acids. These consist of the following seven macro elements: calcium, magnesium, sodium, potassium, phosphorus, chlorine, and sulphur. Additionally, they supply essential trace elements that are necessary for optimal health, such as manganese, iron, copper, zinc, selenium, cobalt, molybdenum, and iodine [8].

With an average yield of 9.96 Mt/ha and an annual production of 1370.21 Mt, India occupied 137.54 (000'ha) of French bean land. West Bengal, Andhra Pradesh, Jharkhand, Jammu and Kashmir, and Himachal Pradesh are the principal French bean-growing states in India. [9]

High-quality seeds are a cornerstone of agricultural productivity, particularly when paired with improved management practices. However, under rainfed conditions, crop yields are often limited by factors such as poor seed quality, soil moisture deficits, erratic rainfall, and suboptimal crop management. Leguminous crops, such as French bean (*Phaseolus vulgaris* L.), face additional challenges due to their hard seed coats, which hinder uniformity and rapid germination, leading to low field emergence and poor crop establishment. This issue is particularly critical in rainfed agriculture, where environmental stresses exacerbate germination problems.

Biofertilizers, which are developed based on living bacteria, fungi, or other microorganisms, offer potential benefits to crops and soil properties. They enhance plant nutrition by facilitating nutrient availability. For instance, nitrogen-fixing bacteria associated with legumes convert atmospheric N₂ into ammonium, which is accessible to plants [10].

They are low-cost, renewable, and eco-friendly sources of plant nutrients that supplement chemical fertilizer. They can be used either in seed treatment or soil application. Bio-fertilizers are also ideal inputs for reducing the cost of cultivation and for practicing organic farming

Use of biofertilizer in crop not only fixes the biological nitrogen but also solubilizes the insoluble phosphates in soil and thus improves fertilizer-use efficiency [11]. These microorganisms play an important role in increasing the availability of N, P and K. Keeping this in view, the present investigation was undertaken to standardize of Different Pre-Sowing Seed Treatment of *Phosphate phosphate-solubilizing bacteria*, *Rhizobium*, *Azospirillum*, and *Trichoderma* on growth, and yield attributing traits of French bean (*Phaseolus vulgaris* L.)

MATERIALS AND METHODS

The Study was conducted at Farm of Genetics and Plant Breeding, SHUATS, Prayagraj. Using a variety (Ellora 11).

Sources of PSB, Rhizobium, Azospirillum, and Trichoderma

Microbial cultures are typically obtained from WESFRA Organic Bio Liquid Manure, Organic Dew's, and Katyayani. The Phosphate-Solubilizing Bacteria (PSB) used in this study is sourced from Organic Dew's Liquid

Pseudomonas fluorescens (2×10⁸ CFU/ml), belonging to the genus *Pseudomonas* (Taxonomy ID: 294) (<https://www.ncbi.nlm.nih.gov/>). The *Rhizobium* culture is obtained from Katyayani's Rhizogen (*Rhizobium* CFU 5×10⁸), under the genus *Rhizobium* (Taxonomy ID: 396) (<https://www.ncbi.nlm.nih.gov/>) and identified as *Rhizobium phaseoli*. The *Azospirillum brasilense* strain used is Organic Dew's *Azospirillum* (2×10⁸ CFU/ml), belongs to the genus *Azospirillum* and species *Azospirillum brasilense* (Taxonomy ID: 192) (<https://www.ncbi.nlm.nih.gov/>). The *Trichoderma* culture is derived from WESFRA Organic Bio Liquid Manure, containing *Trichoderma viride*, which belongs to the genus *Trichoderma* (Taxonomy ID: 5547) (<https://www.ncbi.nlm.nih.gov/>) and species *Trichoderma viride*.

Preparation of Different Doses (8%, 10%, and 12%) of Inoculants

To make different dosages (8%, 10%, and 12%) of the microbial inoculants. *Pseudomonas fluorescens*, at 2×10⁸ CFU/ml, the 8% dose requires 1.6 ml of inoculant in 100 ml of distilled water, equating to a CFU of 1.6×10⁶ /ml, while the 10% dose requires 2 ml, equating to a CFU of 2×10⁶ /ml, and the 12% dose needs 2.4 ml, equating to a CFU of 2.4×10⁶ /ml. *Rhizobium*, 5×10⁸ CFU/ml, the 8% dose consists of 4 ml in 100 ml of distilled water (4×10⁶ CFU/ml), the 10% dose requires 5 ml (5×10⁶ CFU/ml), and the 12% dose needs 6 ml (6×10⁶ CFU/ml). *Azospirillum brasilense* 2×10⁸ CFU/ml the 8% dose would consist of 1.6 ml in 100 ml of distilled water that equates to 1.6×10⁶ CFU/ml; 10% would be 2 ml 2×10⁶ CFU/ml, and 12% would require 2.4 ml that amounts to 2.4×10⁶ CFU/ml. *Trichoderma viride* prepare the 8% dose, 8 ml mix in 100 ml distilled water. The 10% dose requires 10 ml and the 12% needs 12 ml respectively. (Table 1).

Observation such as Field emergence, Plant height at 30&60 DAS, No of primary branches, Days.to.50.. Flowering, No.of.pods/plant, Number.of.seeds/pod, Pod Yield/plant(g), Seed yield/Plant(g), Seed Index(g). Biological.Yield (g), Harvest.Index. were recorded from 5 randomly selected plants

Field Emergence (%) = (Number of seeds sown / Number of seedlings emerged) × 100

Seed Index (g) = Weight of 100 seeds

Harvest Index (%) = (Biological Yield / Seed Yield) × 100

Table 1. List of Pre-sowing seed treatments along with their concentration dose and duration

Treatment Code	Treatment	Dose (%)	Duration
T0	Control (Untreated)	—	—
T1	PSB	12%	6 hours
T2	PSB	10%	6 hours
T3	PSB	8%	6 hours
T4	<i>Azospirillum</i>	12%	6 hours
T5	<i>Azospirillum</i>	10%	6 hours
T6	<i>Azospirillum</i>	8%	6 hours
T7	<i>Rhizobium</i>	12%	6 hours
T8	<i>Rhizobium</i>	10%	6 hours
T9	<i>Rhizobium</i>	8%	6 hours
T10	<i>Trichoderma</i>	12%	6 hours
T11	<i>Trichoderma</i>	10%	6 hours
T12	<i>Trichoderma</i>	8%	6 hours

Statistical analysis

The process outlined [12] was used to statistically analyze the experiment's results utilizing a c randomized design. The software program Grapes 1.1.0 was utilized to determine the crucial differences (CD) values. At a 5% probability level, the crucial differences (CD) were computed and Dunken letters were mounted.

RESULTS

Effect of different pre-sowing seed treatments on morphological traits

Field emergence (%): Significant difference was observed in field emergence, where the highest field emergence was recorded in T7 (88.84%), followed by T6 (87.35%) and T9 (85.43%), all of which were significantly superior to the control T0 (71.17%) (Table 2).

Plant height (cm): Significant difference was observed in plant height. T7 exhibited the tallest plants at 30 days after sowing (24.77 cm), followed by T6 (23.50 cm) and T4 (20.88 cm), with the control showing the shortest plants (19.15 cm). The maximum plant height at 60 DAS was observed in T7 (45.37 cm), followed by T6 (44.37 cm) and T5 (42.38 cm), whereas the control recorded a significantly lower height of 38.28 cm (Table 2).

Number of primary branches: Significant difference was observed in no. of primary branches produced. T7 produced the maximum no. primary branches (4.75), followed by T6 (4.57) and T8 (4.12), while the control showed the lowest number (2.58) (Table 2).

Days to 50% Flowering: Significant difference was observed in plant attaining to 50% flowering. T7 had the earliest flowering (44.00 days), indicating faster reproductive development, followed by T6 (44.33 days) and T3 (45.00 days), compared to the latest flowering in the control at 47.66 days (Table 2).

Effect of different pre-sowing seed treatments on yield traits

Number of pods per plant: Significant difference was observed in no of pods produced in each plant. The maximum number of pods was found in T7 (14.50), followed by T6 (13.26) and T9 (11.38), while the control recorded (7.66) pods (Table 3).

Number of seeds per pod: Significant difference was observed in no of seeds produced by each pod. T7 leads in maximum seed number per pod (5.39), followed closely by T6 (5.21) and T8 (5.17), whereas the control had the lowest (3.24) (Table 3).

Pod Yield per Plant (g): T7 significantly outperformed all other treatments with the highest yield (201.68 g), followed by T6 (174.85 g) and T9 (174.54 g), while the control was the least productive (113.42 g) (Table 3).

Seed Yield per Plot (g): Significant difference was observed in seed yield/plot. The seed yield was highest in T7 (1087.53 g), followed by T6 (910.91 g) and T8 (867.10 g), compared to a low yield in the control (367.99 g) (Table 3).

Seed Index (g): Significant difference was observed in Seed Index. T7 recorded the highest seed index (39.06 g), followed by T10 (37.47 g) and T3 (36.15 g), while T0 showed the lowest value (33.95 g) (Table 3).

Biological Yield (g): Significant difference was observed in biological yield. the highest biological yield was observed in the T7 (120.61 g), followed by T5 (119.09 g) and T9 (119.08 g) (Table 3).

Harvest Index (%): T7 had the highest harvest index (4.15%), indicating superior resource use efficiency, followed by T6 (3.11%) and T8 (3.10%), while the control had the lowest value (1.22%) (Table 3).

This parameter-wise analysis demonstrates the substantial improvement in plant growth, yield attributes, and productivity in French bean through pre-sowing treatment with *Rhizobium* (especially at 12%), clearly outperforming the untreated (Control) seeds.

Table 2. Effect of different pre-sowing seed treatments on morphological traits

Treatment	Field emergence (%)	Plant height @30DAS (cm)	Plant height @30DAS (cm)	No. of Primary branches	Days to 50% Flowering
T0	71.17±1.26g	19.15±0.06e	38.28±0.17f	2.58±0.01i	47.66±1.50a
T1	73.85±1.20fg	19.88±0.13de	41.42±0.67de	2.54±0.08i	46.00±1.87abcd
T2	72.33±3.06fg	21.15±0.42c	41.21±0.37de	3.25±0.11fg	46.00±1.62abcd
T3	75.71±0.61ef	21.43±0.10c	41.03±1.74de	3.17±0.13g	45.00±0.81bcd
T4	83.45±3.01bc	20.88±0.87cd	40.71±1.76e	3.50±0.01e	46.33±1.04abc
T5	78.60±2.83de	21.87±0.16c	42.38±0.27cde	2.94±0.05h	47.66±0.39a
T6	87.35±2.60ab	23.50±1.04b	44.37±1.68ab	4.57±0.17b	44.33±0.28cd
T7	88.84±2.96a	24.77±1.07a	45.37±0.86a	4.75±0.04a	44.00±0.87d
T8	80.65±1.96cd	21.85±0.75c	43.32±0.70bc	4.12±0.00c	45.66±0.78abcd
T9	85.43±2.85ab	21.04±0.19c	40.78±1.10e	3.37±0.14ef	47.33±1.54a
T10	78.59±2.76de	21.46±0.91c	42.81±0.85bcd	3.94±0.00d	47.00±0.04ab
T11	84.27±0.46bc	21.97±0.46c	42.31±0.19cde	3.12±0.06g	45.66±0.49abcd
T12	83.48±2.03bc	21.45±0.85c	41.31±0.82de	3.47±0.00e	46.66±2.06ab

Duncan's multiple range test was calculated at the level of significance ($p < 0.05$), mean values with different letters are significantly different. T₀ - control, T₁ - PSB 12%, T₂ - PSB 10%, T₃ - PSB 8%, T₄ - *Azospirillum* 12%, T₅ - *Azospirillum* 10%, T₆ - *Azospirillum* 8%, T₇ - *Rhizobium* 12%, T₈ - *Rhizobium* 10%, T₉ - *Rhizobium* 8%, T₁₀ - *Trichoderma* 12%, T₁₁ - *Trichoderma* 10%, T₁₂ - *Trichoderma* 8%.

Table 3. Effect of different pre-sowing seed treatments on yield traits

Treatment	No. of pods/plant	No. of seeds/Pod	Pod yield/ Plant(g)	Seed yield/ plot (g)	Seed index (g)	Biological yield (g)	Harvest index
T0	7.66±0.26h	3.24±0.10f	113.42±1.52h	367.99± 6.00h	33.95±0.17e	104.83±0.76e	1.22±0.05h
T1	9.66±0.17g	4.00±0.13e	122.66±0.22g	490.93± 7.97g	34.91±0.63cde	115.13±2.49bcd	1.70±0.06g
T2	10.37±0.31f	4.03±0.07e	126.72±4.57fg	511.17±17.97fg	36.28±1.41bc	114.15±1.44cd	1.79±0.05g
T3	9.60±0.13g	4.11±0.05de	128.43±3.13efg	527.99± 8.09f	36.15±0.91bc	111.33±0.60d	1.90±0.07f
T4	11.46±0.09c	4.06±0.12e	146.38±2.24d	594.54±12.33e	34.32±0.74de	113.62±1.54cd	2.10±0.08e
T5	10.95±0.11de	4.12±0.12de	147.77±4.66d	623.39±16.30e	34.76±0.69cde	119.09±0.21ab	2.09±0.03e
T6	13.26±0.51b	5.21±0.19a	174.85±4.89b	910.91±15.60b	35.78±0.26cd	117.30±2.33abc	3.11±0.03b
T7	14.50±0.24a	5.39±0.02a	201.68±2.91a	1087.53±33.33a	39.06±0.14a	120.61±4.36a	4.15±0.07a
T8	9.93±0.13g	5.17±0.21a	167.50±7.10c	867.10±29.70c	34.79±1.29cde	111.74±1.21d	3.10±0.05b
T9	11.38±0.01c	4.04±0.10e	174.54±3.62b	706.09±28.64d	35.42±1.56cde	119.08±3.43ab	2.37±0.02d
T10	10.70±0.01ef	4.59±0.20b	134.10±5.68e	616.48±17.23e	37.47±0.24b	116.36±4.41abc	2.12±0.09e
T11	10.37±0.08f	4.30±0.09cd	164.18±1.33c	706.13± 5.09d	34.43±0.43de	114.50±0.52cd	2.47±0.04c
T12	11.14±0.07cd	4.51±0.07bc	131.27±0.83ef	592.62±14.96e	35.44±0.29cde	111.57±1.51d	2.12±0.08e

Duncan's multiple range test was calculated at the level of significance ($p < 0.05$), mean values with different letters are significantly different.

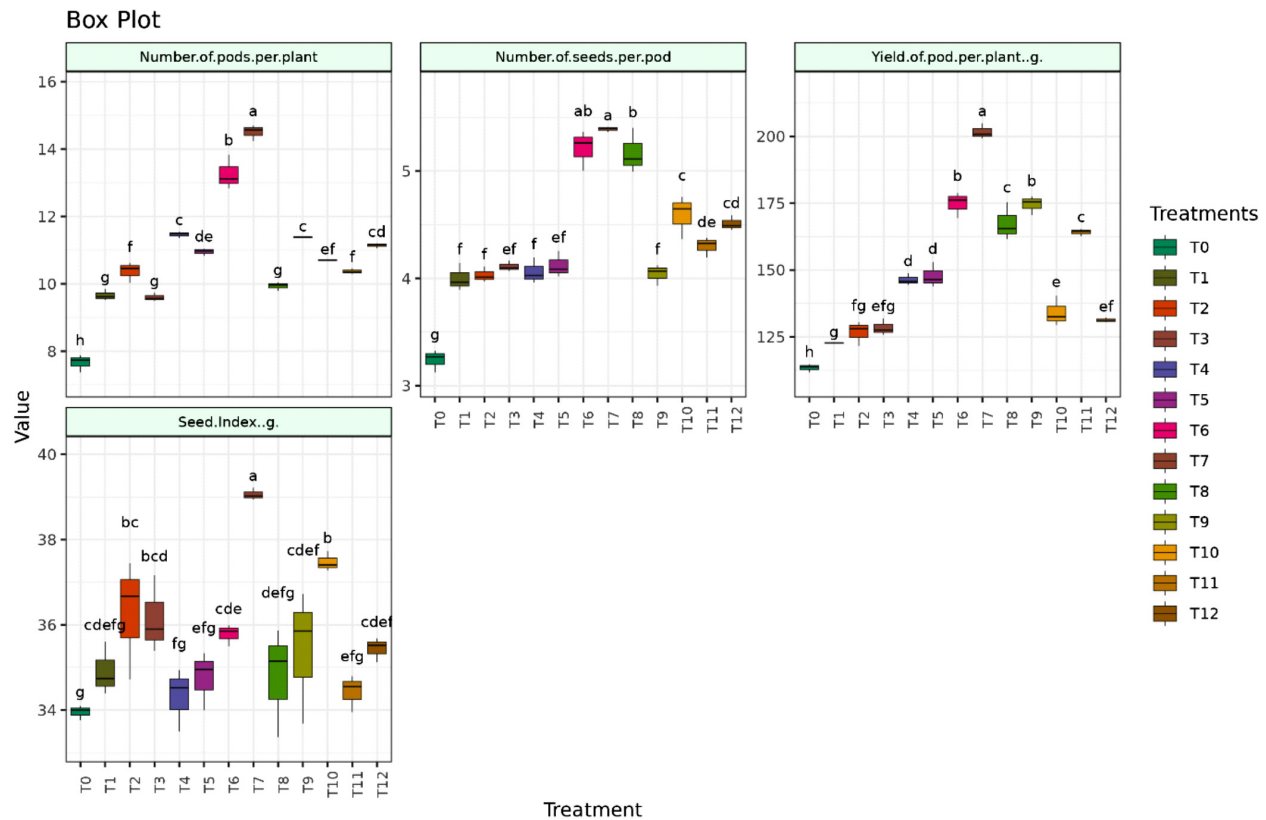
DISCUSSION

Biofertilizers such as *Phosphate Solubilizing Bacteria*, *Rhizobium*, *Azospirillum* and *Trichoderma* improve seed quality by enhancing soil fertility and promoting beneficial microbial activity. They increase the availability of key nutrients (N, P, K), improve soil organic matter, and suppress pathogens, leading to healthier roots and stronger plant growth. This results in better nutrient uptake, improved seed filling, higher seed weight, and overall superior seed quality.

The present study clearly demonstrates the significant impact of *Rhizobium* inoculation on the growth, development, and yield attributes of Frenchbean

(*Phaseolus vulgaris* L., var. Ellora Number 11), with treatment T7 emerging as the most effective among all tested treatments. Consistent improvement was observed across key parameters such as field emergence, plant height, number of primary branches, days to 50% flowering, number of pods per plant, seed yield per plot, and harvest index when compared to the untreated control (T0).

These findings are in agreement with [13], who reported that *Rhizobium* inoculation in French bean significantly enhanced yield due to increased leaf number and plant height. This can be explained through the source-sink relationship, where more photosynthates produced by a larger leaf area are translocated to sink organs such as



T₀ - control, T₁ - PSB 12%, T₂ - PSB 10%, T₃ - PSB 8%, T₄ - *Azospirillum* 12%, T₅ - *Azospirillum* 10%, T₆ - *Azospirillum* 8%, T₇ - *Rhizobium* 12%, T₈ - *Rhizobium* 10%, T₉ - *Rhizobium* 8%, T₁₀ - *Trichoderma* 12%, T₁₁ - *Trichoderma* 10%, T₁₂ - *Trichoderma* 8%.

Figure 1. Representation of various pre-sowing seed treatments using a Box plot for various parameters

Pods and seeds, resulting in greater productivity. Similarly, Rabbani [14] noted a significant increase in harvest index in soybean under 100% *Rhizobium* application, which was attributed to better vegetative growth and effective utilization of nutrients from both organic and inorganic sources. In the present study, a similar trend was observed, where T₇ treatment recorded the highest harvest index (4.15%), likely due to improved plant vigour and seed development.

The beneficial effects of *Rhizobium* on field emergence were evident in our study, with T₇ showing the highest emergence rate (88.84%). This corroborates the findings of Shivangi [15] who observed that *Rhizobium* combined with PGPR significantly improved germination and early seedling establishment in French bean. The poor emergence observed in the control (T₀) might be due to the inability of untreated seeds to effectively mobilize stored reserves during early germination. In contrast, inoculated seeds benefit from improved synthesis of secondary metabolites and plant growth-promoting substances that enhance early vigor.

Vegetative growth traits such as plant height and branching were also enhanced with *Rhizobium* treatment. This is likely due to the production of growth-promoting substances such as auxins, cytokinins, gibberellins, and amino acids, as well as improved nitrogen fixation, all of which contribute to robust shoot and root development [16]. The increased number of primary branches in T₇ (4.75) and improved plant height at both 30 and 60 DAS further support these physiological mechanisms.

Yield components such as the number of pods per plant, seeds per pod, and seed index were also positively influenced by *Rhizobium* inoculation. The highest pod and seed yields recorded in T₇ (201.68 g/plant and 1087.53 g/plot, respectively) highlight the effectiveness of Rlv in improving reproductive efficiency. [16] similarly reported enhanced seed weight in French bean under *Rhizobium* and *Rhizobium* + PGPR treatments, respectively, due to better nutrient assimilation and seed filling.

At a biochemical and molecular level, as observed in *Rhizobium* inoculation has been associated with

increased seed biomass and protein/lipid content, facilitated by enhanced nitrogen and amino acid metabolism [17]. Proteomic studies revealed the induction of Calvin cycle-related proteins like RuBisCO and decreased expression of LEA proteins, indicating a shift toward rapid development and altered hydration dynamics in seeds [18]. Although our study did not include proteomic analysis, the improved seed quality and seed index under T7 (39.06 g) may suggest similar underlying physiological processes.

CONCLUSION

Based on the findings of the present study, it can be concluded that pre-sowing seed treatment of French bean (variety Ellora Number 11) with *Rhizobium* @12% for 6 hours (T7) significantly improved field emergence, vegetative growth, and yield attributes, followed closely by *Azospirillum* @8% for 6 hours (T6), when compared to untreated seeds. *Rhizobium* inoculation not only enhanced plant height, branching, and pod yield, but also improved seed quality through better nutrient assimilation and nitrogen fixation. The application of biofertilizers also contributed to enriched soil health, improved root development, and reduced dependence on chemical inputs. Therefore, biofertilizer-based seed treatments are an eco-friendly, cost-effective, and sustainable approach to boosting productivity in French bean cultivation. These treatments promote uniform crop establishment, reduce re-seeding and input costs, and support the production of safer, residue-free food beneficial for both consumers and the environment.

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