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Field evaluation of biostimulants on growth, flowering, yield, and quality of snap beans in subtropical environment

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Abstract: The cultivation of snap beans (*Phaseolus vulgaris* L.) in subtropical regions faces environmental challenges leading to potential declines in yield. This study explores the efficacy of biostimulants as a solution, specifically investigating spraying treatments with 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) on the snap bean cv. Paulista. Over two growing seasons with late sowing and elevated summer temperatures, the research assesses growth, flowering, yield, and quality. Notably, 5 ppm TRIA demonstrates the most significant impact on plant growth and leaf nutrient content. Treatments with 40 ppm 6-BA, 5 ppm TRIA, or 200 ppm KSi exhibit notable effects on inflorescence flower count and flowers per plant. These treatments prove most effective for crucial green pod yield measures, including the number and weight of marketable pods. Moreover, 40 ppm 6-BA or 5 ppm TRIA significantly enhances pod characteristics, such as length, diameter, and weight, consistently improving over both seasons. Particularly, 5 ppm TRIA outperforms in enhancing the chemical quality of pods throughout the study. Overall, the findings suggest that the application of 5 ppm TRIA offers the most favorable enhancements for the growth, flowering, productivity, and quality of snap bean plants in subtropical field conditions.

1. Introduction

The global population is now and will continue to exert increased pressure on the need for food. Hence, it is essential for farmers to annually increase food production with the existing resources to fulfill this demand. Common bean (*Phaseolus vulgaris* L.) is a common vegetable that includes both snap and dry beans (Lin *et al.*, 2008). In Egypt, farmers dedicate 27363 hectares to green bean cultivation, yielding 284299 tons annually (FAOSTAT, 2021).However, snap beans have a notable susceptibility to high summer temperatures, particularly when subjected to delayed planting, like in April and May under Egyptian field conditions (El-Bassiony *et al.*, 2012). While it has been stated that the optimal temperature for bean plants is 23°C (Dickson and Boettger, 1984). Omae *et al.* (2006) observed that the occurrence of high summer temperatures (26°C Min and 30°C Max.) during the initiation of the blooming stage had an adverse impact on the quantity and the weight of pods. Along with this, climate models predict a 50% decrease in global cultivated area by 2050 due to global warming (Rippke *et al.*, 2016; Rama Rao *et al.*, 2022).

One potential approach to enhancing snap bean production is through the breeding of new cultivars. However, it is important to note that this process often takes a significant amount of time and may provide limited results (Xiong *et al.*, 2022). Biostimulants provide a compelling alternative in the context of degraded agricultural regions and the risks associated with climate change. In recent times, there has been increased research focusin the utilization of biostimulantsin the form of plant growth regulators (e.g., 6-benzylaminopurine; 6-BA as a synthetic cytokinin, and triacontanol; TRIA), chitosan (Ch), and trace elements (e.g., silicon; Si) that have been found effective in improving plant productivity (Du Jardin, 2015; Yaghubi *et al.*, 2019; Islam and Mohammad, 2020; Hassan *et al.*, 2021; Stasińska-Jakubas and Hawrylak-Nowak, 2022). Although cytokinins (CKs) have vital function in controlling plant development, they have also been shown to confer other benefits, such as improving photosynthetic rates, photosynthetic pigments, and nutrient uptake (Aremu *et al.*, 2020; Li *et al.*, 2021). In a study conducted by Mostafa and Brengi (2018), it was shown that the application of 6-BA solution on okra leaves resulted in improved yield and chemical composition. Furthermore, Yang *et al.* (2016) illustrated that the treatment with 6-BA resulted in an improvement in several aspects of wheat grain development, including wheat grain filling and endosperm cell division under heated growth conditions. It is a widely recognized that TRIA is plant growth regulator (Islam and Mohammad, 2020). Triacontanol is a saturated alcohol initially discovered in alfalfa (Ries *et al.*, 1977) and is found naturally as a wax coating on a variety of plant species (Islam and Mohammad, 2020). In addition to its function in eliciting responses to stresses, TRIA is participated in plant growth, production, and vital physiological processes (Faiz *et al.*, 2024). In this manner, Waqas *et al.* (2016) showed that both normal growth and heat stress conditions, TRIA treatment of mung bean plants resulted in improved plant growth, leaf chlorophyll content, nutrients, and protein content. Chitosan is a naturally carbohydrate polymer that has been produced from chitin, a substance found in the shells of crustaceans (Hidangmayum *et al.*, 2019). It is non-toxicand biocompatible, making it potentially useful in agriculture and biotechnology (Stasińska-Jakubas and Hawrylak-Nowak, 2022). Basically, Ch improves physiological responses and reduces the negative effects of abiotic stressors through the secondary messengers (Hidangmayum *et al.*, 2019). Therefore, Ch is thought to be a viable exogenous addition for increasing crop production and overcoming abiotic stress (Stasińska-Jakubas and Hawrylak-Nowak, 2022). Apart from this, Ch also enhanced the productivity of many crops such as tomatoes (El-Tantawy, 2009), cowpea (Farouk and Amany, 2012) and cucumber (Ali *et al.*, 2020). Generally, silicon (Si)ranks among the most abundant elements found in soil (Souri *et al.*, 2021). Recently, the connections between Si and various biological processes in multiple crops were clarified, and silicon was recognized as one of the vital nutrients required by plants (Zargar *et al.*, 2019). Silicon is engaged in many biological activities such as photo synthesis, nutrient uptake, and plant adaptation to stress (Zargar *et al.*, 2019; Souri *et al.*, 2021). Potassium silicate (KSi) is usually used as biostimulant and a producer of both soluble K and Si (Yaghubi *et al.*, 2019). It is well recognized that K is a core element and participates in a vital function in cell division, protein synthesis, the formation of sugars, and plant growth, as well as vital processes such as plant photosynthesis and stomata movement (Ali *et al.*, 2021).

Although previous studies have examined the individual impacts of these elicitors on plant growth, a comprehensive investigation into their effects specifically on snap bean plants remains lacking. Moreover, these studies evaluated different parameters and were conducted in different growing environments; consequently, the field evaluation of these biostimulants under a particular subtropical summer conditions are required. Thus, this research was created to test the beneficial impacts of 6-BA, Ch, TRIA, or KSi on the growth, blooming, productivity, and quality of snap bean plants grown in delayed summer cultivation in a subtropical environment.

2. Materials and Methods

Snap bean field conditions

Field trials were undertaken in the Sidi Ghazy Region of Kafr El-Dawar city, located in the Beheira Governorate of Egypt. These experiments were done during the seasons of 2021 and 2022. The geographical coordinates of the study area are around 31°07'N latitude and 30°08' E longitude. The region has an arid climatic condition characterized by an annual precipitation of about 90-110 mm, mostly in the form of ineffectual showers during the winter. Figure 1 presents a summary of the monthly temperatures, measured over the course of two cultivation seasons. The source of this data originates from the Egyptian Ministry of Agriculture and Reclamation of Soils, bulletin of agricultural meteorological data. Samples from the trial soil were subjected to drying and then sifted by a two-mm sieve. These samples were then analyzed using the protocols outlined by Page *et al.* (1982). Experimental soil had a clay texture (22.5% sand, 35.4% silt, and 42.1% clay) with a pH of 8.14, EC value of 1.19 dsm^{-1}, and an organic material level of 1.75%, as an average over the two seasons.

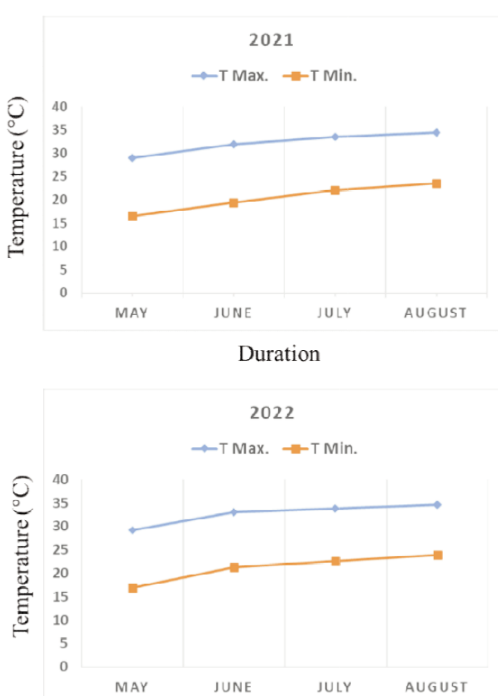

Fig. 1 - Monthly means of maximum (T Max.) and minimum (T Min.) temperature during the two studied seasons (summer 2021 and 2022) (Egyptian Ministry of Agriculture and Reclamation of Soils, bulletin of agricultural meteorological data).

Plant material, experimental design, and treatments

Seeds of the snap bean (*Phaseolus vulgaris* L.) cv. Paulista (Alsafwa company, Egypt) were planted on May 1st and May 2nd in the seasons of 2021 and 2022, respectively. The selection of this cultivar was based on its significant economic worth in both local and foreign markets. The experimental site was plowed and leveled adequately before plots were established in accordance with the experimental layout. The experimental treatments were organized as stated by the Randomized Complete Block Design (RCBD). The trial consisted of 9 treatmentswith3replicates (plots). The seeds were manually cultivated on a single side of the ridge, with a spacing of 10 cm between each seed. The ridge itself had a dimension of 60 cm and 5 m long. Each plot consisted of three ridges, and the total size of each plot was 9 m^2.Two guard ridges were present between each treatment to prevent spray drift.

In this investigation, four biostimulants plus a control were examined, namely 6-benzylaminopurine (6-BA, Sigma-Aldrich, USA) at 20 and 40 ppm, chitosan (Ch, Sigma-Aldrich, USA) at 100 and 200 ppm, triacontanol (TRIA, Sigma-Aldrich, USA) at 2.5 and 5 ppm, potassium silicate (KSi, Sigma-Aldrich, USA) at 100 and 200 ppm, and a control group treated with distilled water. Former studies were employed to identify the suitabledose range of 6-BA (Abouelsaad and Brengi, 2022; Zarea and Karimi, 2023) Ch (Ibrahim and Ramadan, 2015; Stasińska-Jakubas and Hawrylak-Nowak, 2022), TRIA (Islam and Mohammad, 2020), and KSi (Ibrahim *et al.*, 2020). Foliar treatments were employed 3 times, 15 days after seed sowing, followed by further applications every 15 days afterwards. The plants were subjected to a foliar application employing a knapsack sprayer throughout the afternoon until drop-off.

All treatments were provided with an equal total quantity of 50 N, 60 P, and 60 K (kg ha^{-1}) fertilizer for the duration of the season. These fertilizers(surface broadcast application) were given in two equal portions, with the first dosage delivered during the third week after seed planting and the second dose applied during the seventh week. Nitrogen, P, and K were provided as ammonium nitrate, single superphosphate, and potassium sulfate, respectively. All the required practices for cultivating snap beans were properly conducted as required.

Plant growth, flowering, chlorophyll content and mineral analysis

After 50 days of planting, three plants from each replicate (nine from each treatment) were taken for measuring the height (cm), branch number (plant^{-1}), and number of leaves (plant^{-1}). The relative content of chlorophyll in snap bean were assessed using the Chl meter instrument (SPAD-502) manufactured by Konica Minolta Sensing in Japan. The assessment included measuring the number of inflorescences plant^{-1}, number of flowers in the inflorescence, total number of flowers plant^{-1}, and evaluating the length of the inflorescence (cm) (at 50% flowering according to Schwartz and Langham, 2010) The fresh weight (g) of shoot was measured, and subsequently, the plants were subjected to oven-drying at a temperature of 60°C till their weights stabilized, leading to the determination of the dry weight (g) of the shoot. Plant leaves area (cm^2) was conducted using a mathematical analysis that examined the relationship between the dry weight of leaves (plant^{-1}) and the dry weight and area of 20 discs taken from fresh leaves using a borer with a known diameter. Wallace and Munger (1965) established this relationship and presented it in the following formula:

$$\text{Leaves area (cm}^2\text{)} = \text{leaves dry weight (g)} \times 20 \text{ discs area (cm}^2\text{)}/20 \text{ discs dry weight (g)}$$

Micro-Kjeldahl was employed to assess Nin snap bean leaves (Sáez-Plaza *et al.*, 2013), but P level was quantified by colorimetric techniques (Watanabe and Olsen, 1965). Zinc, Fe, Mg, and Mn were analyzed in leaves using the atomic absorption spectrophotometer model Perkin Elmer 3100, while K and Ca levels of leaf tissue were quantified by a flame-photometer (CORNING M410) as employed by Munns *et al.* (2010).

Green pod yield and quality

At harvest time (65 days from sowing), observations were recorded for seven characters *viz.,* the number of pods (plant^{-1}), the weight of pods (g plant^{-1}), the number of marketable pods (plant^{-1}), the weight of marketable pods (g plant^{-1}), the mean of pod weight (gm), pod length (cm) and pod diameter (mm). This research considers pods that possess significant quality traits that are important for the export market, such as well-formedness, uniformity, straightness, and absence of flaws, as being considered marketable. Total N (%) was determined in green pods using the micro-Kjeldahl apparatus as defined by Sáez-Plaza *et al.* (2013). Subsequently, total protein (%) was calculated using N% (Mariotti *et al.*, 2008). The detection of vitamin C in the green pods was carried out at a wavelength of 525 nm, utilizing the methodology previously established by Srivastava and Singh (1988). To establish a standard curve, the utilization of ascorbic acid (Analytical Reagent, Solarbio) was employed. The quantification of vitamin C is presented in mg100 g^{-1} FW. The measurements of crude fiber and soluble sugar in pods were conducted using the methodology established by Slavin (1987) and Rady *et al*. (2019), respectively.

Data analysis

The data underwent statistical analysis using a one-way factorial design within the framework of a Randomized Complete Block Design (RCBD). The COSTAT program (CoStat program version 6.311, 2005) was used to conduct a statistical analysis, namely the Duncan's multiple range test, with a significance threshold of P≤0.05, for the purpose of comparing the means.

3. Results and Discussion

Snap bean growth and chlorophyll contents

Changes in snap bean growth parameters caused by the foliar application of growth elicitors (6-BA, Ch, TRIA, or KSi) are presented in Tables 1 and 2. The application of all treatments boosted plant growth, as clarified by the increases in snap bean height, leaves area, shoot fresh and dry weights. In most instances, the applied treatments also resulted in boosted the number of leaves and branches, although in the case of the treatment with 100 ppm Ch, both seasons' values were similar to the control treatment. Several studies have provided evidence suggesting that the applications of 6-BA, Ch, and KSi have the potential to increase the overall snap bean growth (Werner and Schmülling, 2009; Hidangmayum *et al*., 2019; Yaghubi *et al*., 2019; Gomaa *et al*., 2021). Cytokinins (CKs) are often characterized as hormones that stimulate growth; however, it should be noted that several substances exhibiting CK activity have been discovered to control many features of plant development (Haberer and Kieber, 2002). Cytokinins influence cell multiplication, that in turn influences plant development, and they also promote adventitious buds growth (Kieber and Schaller, 2014). Such substances, including exogenous applications, were used to promote growth in crops and vegetables (Yang *et al*., 2016; Mostafa and Brengi, 2018; El-Areiny *et al*., 2019; Aremu *et al*.,

Table 1 - Plant height, shoot fresh weight, and shoot dry weight of snap beanas affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	Plant height (cm)		Shoot fresh weight (g)		Shoot dry weight (g)	
	2021	2022	2021	2022	2021	2022
Control	44.30 e	45.63 d	293.07 d	301.90 d	29.83 d	30.89 e
6-BA (20 ppm)	50.33 bc	52.33 ab	326.02 c	339.03 c	35.64 bc	37.30 cd
6-BA (40 ppm)	52.30 a	53.83 a	364.40 ab	375.09 ab	37.79 b	39.04 bc
Ch (100 ppm)	48.43 cd	49.93 c	351.40 b	361.91 bc	34.77 c	35.99 d
Ch (200 ppm)	47.97 d	51.67 bc	357.60 b	385.00 ab	35.48 bc	38.66 bc
TRIA (2.5 ppm)	49.67 cd	52.80 ab	355.13 b	377.60 ab	36.62 bc	39.31 bc
TRIA (5 ppm)	51.90 ab	53.53 a	373.93 a	386.23 a	41.31 a	42.84 a
KSi (100 ppm)	48.17 d	52.17 ab	355.83 b	385.54 a	36.46 bc	40.00 b
KSi (200 ppm)	50.10 bc	53.93 a	360.27 ab	387.89 a	35.94 bc	39.16 bc

Means with different letters for each plant parameter are considered significantly different ($p<0.05$) using the Duncan's multiple range test.

Table 2 - The number of branches, number of leaves, and leaves area of snap beans affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	No. of branches plant^{-1}		No. of leaves plant^{-1}		Leaves area (cm^2)	
	2021	2022	2021	2022	2021	2022
Control	6.67 c	6.67 b	20.00 f	20.67 c	2712.00 e	2793.67 e
6-BA (20 ppm)	7.33 abc	7.33 ab	21.33 cde	22.33 b	2901.33 d	3017.00 cd
6-BA (40 ppm)	8.33 a	8.33 a	23.33 a	24.00 a	3144.67 a	3292.67 a
Ch (100 ppm)	7.00 bc	7.00 ab	20.33 ef	20.67 c	2845.00 d	2930.33 de
Ch (200 ppm)	7.33 abc	8.00 ab	21.00 def	22.67 ab	2992.00 c	3222.00 ab
TRIA (2.5 ppm)	7.67 abc	8.33 a	21.67 bcd	23.33 ab	3096.67 ab	3237.33 ab
TRIA (5 ppm)	8.00 ab	8.33 a	22.67 ab	23.67 ab	3130.67 a	3230.67 ab
KSi (100 ppm)	7.67 abc	8.33 a	21.33 cde	23.33 ab	2886.67 d	3127.00 bc
KSi (200 ppm)	7.67 abc	8.33 a	22.33 abc	24.00 a	3018.00 bc	3250.00 ab

Means with different letters for each plant parameter are considered significantly different ($p<0.05$) using the Duncan's multiple range test.

2020; Abouelsaad and Brengi, 2022). Additionally, Ch, a biopolymer, employed in crops production primarily owing to its biocompatible and biodegradable nature, together with its notable biological activity (Hidangmayum *et al*., 2019). Despite not being a constituent of plant tissues, Ch significantly boosts the development and growth of plants (Stasińska-Jakubas and Hawrylak-Nowak, 2022). This was confirmed by El-Miniawy *et al*. (2013), who claimed that spraying Ch increased the growth (height, leaf area, and weight) of strawberry cv. Sweet Charlie. In another study, foliar application of Ch increased both the growth and nitrate reductase activity of okra (Mondal *et al*., 2012). Recently, ample evidence has shown that Si is a key nutrient for crops such as grains, legumes, and vegetables (Souri *et al*., 2021). Both *in vitro* and field investigations confirmed the favorable benefits of Si in boosting plant development, especially in stressful situations (Zargar *et al*., 2019; Souri *et al*., 2021). According to Eneji *et al*. (2008), Si has been shown to act as a bioregulator and have the capacity to enhance plant development. The usage of KSihas shown a substantial influence on the growth of several agricultural crops such as maize and strawberry (Yaghubi *et al*., 2019; Ibrahim*et al*., 2020; Gomaa *et al*., 2021).

Nevertheless, within the range of applied treatments, it was observed that the application of 5 ppm TRIA had a more pronounced impact on plant growth over both seasons. As average for the two growing seasons, with 5 ppm TRIA the snap bean height boosted by 17.23%, the leaf area expanded by

15.54%, the snap bean fresh weight boosted by 27.76%, and the shoot dry weight boosted by 38.58% as compared to the control. Triacontanol (TRIA) is plant growth regulator that has a significant function in facilitating many plants metabolic processes, ultimately resulting in enhanced growth and development (Naeem *et al.*, 2012; Islam and Mohammad, 2020). Its foliar application at low concentrations stimulates the plant biomass of the crops under both control and stressful circumstances (Naeem *et al.*, 2012). A growing body of research has shown that TRIA is an important factor in controlling a wide range of plant morphological responses. One notable effect is its ability to promote many aspects of plant growth, such as increased height, enhanced biomass, greater leaf number, and expanded leaf area across multiple crop species (Naeem *et al.*, 2012). This increase in plant growth might be because TRIA activates L (+)-adenosine, a second messenger that sends signals throughout the plant to boost growth by promoting cell expansion and proliferation (Masroor *et al.*, 2006; Naeem *et al.*, 2012).

The growth of plants is greatly impacted by the level of photosynthetic pigments, that is critical for photosynthesis. In the current study, the use of spraying treatments has resulted in enhancements in chlorophyll contents, but these improvements were seen at comparable levels in most instances (Table 3). Studies have also shown the effect of CKs, Ch, TRIA, orKSi on increasing the content of photosynthetic pigments. Cytokinins can impede or decelerate the process of plant senescence by inhibiting the degradation of chlorophyll, hence preserving the green color of the leaves (Werner and Schmülling, 2009; Kieber and Schaller, 2014). Meanwhile, treating wheat leaves with 6-BA has been shown to boost the production of the chlorophyll founder, D-aminolevulinic acid (Wang *et al.*, 2022). Also, the spray of Ch has been reported to boost the levels of photosynthetic pigments in rice plants (Pongprayoon *et al.*, 2013) and creeping bentgrass plants suffering temperature stress conditions (Huang *et al.*, 2021). In another study, TRIA shown a notable increase in pigment content, namely chlorophyll a, b, and carotenoids, by 25.6, 33.9, and 13.0% respectively, in the leaves of basil plants, relative to the control (Hashmi *et al.*, 2011). Also, Masroor *et al.* (2006) showed similar results in their study, where they noted a substantial rise in chlorophyll and carotenoid content in tomato seedlings that were treated with TRIA. Former research has also verified the beneficial influence of KSi on the chlorophyll levels in plant leaves (Yaghubi *et al.*, 2019; Zargar *et al.*, 2019; Tejada-Ruiz *et al.*, 2020; Gomaa *et al.*, 2021).

Table 3 - Relative chlorophyll content (SPAD value) of snap bean as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	Relative chlorophyll content (SPAD value)	
	2021	2022
Control	41.33 c	41.67 c
6-BA (20 ppm)	41.67 bc	43.00 b
6-BA (40 ppm)	42.67 a	43.67 ab
Ch (100 ppm)	41.67 bc	43.33 ab
Ch (200 ppm)	42.33 ab	43.67 ab
TRIA (2.5 ppm)	42.67 a	43.33 ab
TRIA (5 ppm)	43.00 a	44.00 a
KSi (100 ppm)	42.67 a	43.00 b
KSi (200 ppm)	42.67 a	43.67 ab

Means with different letters for each plant parameter are considered significantly different ($p<0.05$) using the Duncan's multiple range test.

Elemental analysis

Nutrients are fundamental for the growth and productivity of agricultural crops. They are needed in varying quantities and play key functions in various biological processes. The application of 6-BA, Ch, TRIA, or KSi contributed to a higher level of macronutrients and micronutrients in snap bean leaves, with some exceptions (Tables 4 and 5). For instance, the treatments with 100 ppm Ch during the first season, 200 ppm Ch during the second season, and 100 ppm Si throughout both seasons demonstrated a P level comparable to that of the control. Additionally, the application of Ch at concentrations of 100 and 200 ppm resulted in limited changes to the Ca and Zn content of the leaves. Previous studies have also documented the positive effects of CKs and Si on the content and uptake of essential nutrients. Cytokinins regulate the plants' capacity to uptake various elements, like N, P, and K (Argueso *et al.*, 2009). In a study conducted by Abouelsaad and Brengi (2022), the application of CKs through foliar means led to a rise in the N and P levels in potato leaves, relative to the control. Haberer and Kieber (2002) reported that CKs regulate the expression of multiple transporter genes, thereby influencing the plant's ability to uptake nutrients. From this perspective, some studies also showed that Si treatment boosts macronutri-

Table 4 - Nitrogen (N), phosphorus (P), potassium (K), and calcium (Ca) of snap bean leavesas affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	N (%)		P (%)		K (%)		Ca (%)	
	2021	2022	2021	2022	2021	2022	2021	2022
Control	3.20 g	3.17 g	0.48 f	0.50 e	2.67 e	2.69 f	2.08 f	2.12 d
6-BA (20 ppm)	3.30 f	3.25 f	0.54 b	0.53 bcd	2.86 d	2.81 e	2.15 def	2.17 c
6-BA (40 ppm)	3.40 bc	3.36 c	0.58 a	0.56 a	3.06 b	2.99 c	2.27 bc	2.22 b
Ch (100 ppm)	3.31 ef	3.29 e	0.49 ef	0.48 f	2.89 d	2.92 d	2.08 f	2.12 d
Ch (200 ppm)	3.37 cd	3.36 c	0.51 cde	0.51 e	2.92 cd	2.94 cd	2.12 ef	2.10 d
TRIA (2.5 ppm)	3.43 b	3.42 b	0.51 cde	0.53 cd	3.10 b	3.16 ab	2.31 b	2.33 a
TRIA (5 ppm)	3.48 a	3.47 a	0.53 bc	0.55 ab	3.25 a	3.21 a	2.40 a	2.36 a
KSi (100 ppm)	3.31 f	3.33 d	0.50 def	0.52 de	2.95 cd	2.93 cd	2.19 de	2.22 b
KSi (200 ppm)	3.36 de	3.38 c	0.52 bcd	0.54 abc	3.02 bc	3.11 b	2.21 cd	2.24 b

Means with different letters for each plant parameter are considered significantly different (p<0.05) using the Duncan's multiple range test.

Table 5 - Manganese (Mn), iron (Fe), zinc (Zn), and manganese (Mn) of snap bean leavesas affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	Mg (%)		Fe (%)		Zn (%)		Mn (%)	
	2021	2022	2021	2022	2021	2022	2021	2022
Control	0.36 e	0.39 g	127.67 d	122.33f	37.33 e	40.33 e	48.33 d	50.00 e
6-BA (20 ppm)	0.41 d	0.43 f	151.33 ab	146.33 c	48.00 b	50.67 b	51.00 cd	52.67 d
6-BA (40 ppm)	0.49 ab	0.47 cd	159.00a	154.67 a	54.00 a	52.00 ab	55.33 ab	56.00 a
Ch (100 ppm)	0.44 cd	0.45 e	137.00 cd	135.00 e	41.00 de	41.67 de	51.33 c	50.00 e
Ch (200 ppm)	0.48 abc	0.49 bc	141.00 c	139.00 de	43.00 cd	42.00 de	55.67 a	54.00 abcd
TRIA (2.5 ppm)	0.48 ab	0.49 bc	155.67 a	152.33 ab	48.67 b	50.33 b	52.33 c	53.00 cd
TRIA (5 ppm)	0.51 a	0.53 a	160.67 a	156.67 a	53.67 a	54.33 a	57.00 a	55.33 ab
KSi (100 ppm)	0.45 bc	0.47 de	139.33 c	142.00 cd	43.00 cd	44.00 cd	52.67 bc	53.67 bcd
KSi (200 ppm)	0.49 a	0.50 b	145.67 bc	147.00 bc	47.00 bc	45.33 c	56.67 a	55.00 abc

Means with different letters for each plant parameter are considered significantly different (p<0.05) using the Duncan's multiple range test.

ent (e.g., P, K, and Ca) and micronutrient (e.g., Cu, and Fe) absorption in crops (Zargar *et al*., 2019).

The results also showed that 5 ppm TRIA significantly raised the average contents of N (9.10%), P (10.2%), K (32.73%), Ca (13.33%), Mg (38.66%), Zn (39.06%), Fe (26.8%), and Mn (14.23%), compared to the control, throughout the two successive growing seasons (Tables 4 and 5). Notably, this treatment was the most effective among the spraying treatments for all the examined nutrients. As previously reported, the use of TRIA demonstrated a significant influence on the levels of N, P, and K in some crops (Masroor *et al*., 2006; Naeem *et al*., 2012; Islam and Mohammad, 2020). Despite limited research on the impact of TRIA on micronutrient content, it may be inferred that TRIA induces modifications in plants, resulting in changed nutrient contents. In a manner similar to the 5 ppm TRIA treatment, the application of 40 ppm of 6-BA revealed the highest content of nutrients, but only for P, Fe, Zn, and Mn (Tables 4 and 5).

Flowering characteristics

Flowering characteristics (e.g., number of flowers and inflorescences) can have a great influence on the productivity of crops. In this study, the applied treatments had a beneficial effect on the length of the inflorescence in comparison to the control treatment, and the 40 ppm 6-BA treatment achieved the highest value in both seasons (Table 6). Additionally, except for 100 ppm Ch (first season) and 20 ppm 6-BA (second season), the foliar treatments had a stimulating impact on the number of inflorescences plant^{-1} (Table 6). While there is less documentation on the impact of Ch and KSi on promoting vegetable flowering, it has been shown to have positive effects on flower crops (Pichyangkura and Chadchawan, 2015; Amer, 2020). Among the applied treatments in this study, the use of 40 ppm 6-BA, 5 ppm TRIA, and 200 ppm KSi resulted in the most significant increase in the number of flowers in the inflorescence and number of flowers plant^{-1}, a trend that persisted over both seasons. Several studies have shown the role of CKs as pivotal regulators of inflorescence morphology in plants, primarily through regulating meristem activity (Kieber and Schaller, 2014). According to D'Aloia *et al*. (2011), flowering is induced in Arabidopsis plants by exogenous CKs applied during non-inductive short days. Similar findings were reported by Rylott and Smith (1990), who demonstrated that synthetic CKs enhance plant productivity and promote competition between generative and vegetative organs.

In the current study, the number of flowers in the inflorescence and the number of flowers plant^{-1} exhibited respective increases of 22.8% and 50.15% in snap bean treated with 5 ppm TRIA, relative to the control treatment (Fig. 2). The application of TRIA has been found to exert a positive influence on the flowering process of various crops. This was confirmed by Baba *et al*. (2017), who clarified that TRIA raised the number of flowers plant^{-1} while also influencing the timing of flowering in strawberry cv. Camarosa. In

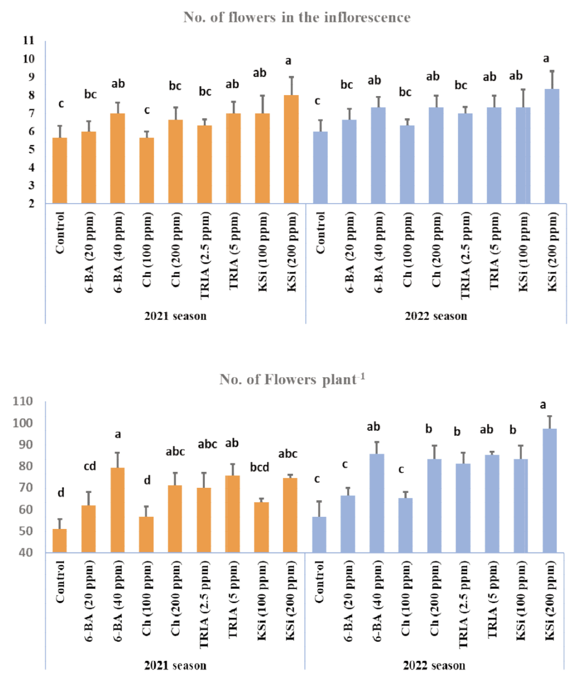

Fig. 2 - The number of flowers in the inflorescence and number of flowers plant^{-1} of snap bean as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the two studied seasons (summer 2021 and 2022). Means (bars) with different letters for each season are considered significantly different (p<0.05) using the Duncan's multiple range test. Data are mean value ± SE.

addition, Sharma *et al.* (2011) tested the effects of TRIA on olives and found that it enhanced the number of flowers in relation to the control. However, some treatments, such as the application of 100 ppm Ch and 20 ppm 6-BA, did not have any notable influence on the number of flowers for the inflorescence

Table 6 - Number of inflorescences and length of inflorescenceof snap bean as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	No. of inflorescences plant^{-1}		Length of inflorescence (cm)	
	2021	2022	2021	2022
Control	9.00 c	9.33 c	7.50 f	7.96 f
6-BA (20 ppm)	10.33 ab	10.00 bc	10.63 cd	10.87 c
6-BA (40 ppm)	11.33 a	11.67 a	13.53 a	12.93 a
Ch (100 ppm)	10.00 bc	10.33 b	8.83 e	9.53 de
Ch (200 ppm)	10.67 ab	11.33 a	9.83 d	9.93 d
TRIA (2.5 ppm)	11.00 ab	11.67 a	10.83 c	11.20 c
TRIA (5 ppm)	11.33 a	11.67 a	12.43 b	11.77 b
KSi (100 ppm)	10.33 ab	11.33 a	8.87 e	9.30 e
KSi (200 ppm)	11.33 a	11.67 a	9.83 d	9.66 de

Means with different letters for each plant parameter are considered significantly different (p<0.05) using the Duncan's multiple range test.

and number of flowers plant^{-1}in relation to the control plants in both cultivation seasons (Table 6).

Snap bean yield

The value of crop yield is determined by the marketable yield, which is a crucial indicator of agricultural productivity. In this study, relative to the control, it was noted that all the evaluated treatments had a positive impact on the total pod number plant^{-1}, except the treatments with Ch at 100 ppm or KSi at 100 ppm for only the first season (Table 7). Also, the implemented treatments caused a significant increase in the number of marketable pods plant^{-1}, the weight of total fresh pods plant^{-1}, and the weight of marketable pods plant^{-1}.

Among the treatments used, applying 6-BA at a concentration of 40 ppm was the most effective treatment to achieve the highest yield parameters, a tendency that held across both seasons. This caused an increase in the total pod number by 36.67%, the number of marketable pods by 49.97%, the weight of total fresh pods by 38.23%, and the weight of marketable pods by 49.49% compared to the control treatment (Table 7). According to Jameson and Song (2016), an elevated concentration of CK throughout the developmental stages of pods and seeds has been identified as a constraining factor in their growth and maturation. A study by Nonokawa *et al.* (2012) also illustrated that CKfor both lupin and soybean crops stopped flower abortion and improved pod set, which ultimately led to a higher yield. Nonetheless, the current data showed that the treatments with 200 ppm Ch, 5 ppm TRIA, and 200 ppm KSi had comparable outcomes to the 40 ppm 6-BA treatment in terms of the number and weight of pods suitable for sale in both seasons (Table 7).

Considering the data shown in Table 8, except for the application of 6-BA at 20 ppm and KSi at 200 ppm during the first season, all treatments exhibited enhancement in the weight of the pods. The application of 6-BA (20 or 40 ppm) and TRIA at 5 ppm, resulted in statistically significant increases in pod length in both growing seasons. Furthermore, the application of Ch at 200 ppm resulted in a notable enhancement of the pod diameter during both growing seasons. Moreover, the implementation of 6-BA (20 or 40 ppm), TRIA (2.5 or 5 ppm), and KSi (100 or 200 ppm) exhibited a significant increase in pod diameter, specifically during the first growing season (Table 8). Similar studies have shown strong evidence supporting the efficacy of Ch, TRIA, and KSi applications for improving the yield and yield components of both vegetable and grain crops (Artyszak, 2018; Kocięcka and Liberacki, 2021).

Green pod quality

The value of yield quality extends beyond mere productivity. It encompasses economic, environmental, social, and health aspects, making it a crucial factor for agricultural production (Abouelsaad *et al*., 2022). As shown in Table 9, the effects of treatments on the quality features (ascorbic acid, fiber, soluble

Table 7 - Number of total pods, number of marketable pods, fresh pods weight, and marketable pods weight of snap bean as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	No. of total pods plant^{-1}		No. of marketable pods plant^{-1}		Fresh pods weight (g plant^{-1})		Marketable pods weight (g plant^{-1})	
	2021	2022	2021	2022	2021	2022	2021	2022
Control	19.00 f	17.67 e	9.43 e	9.33 d	100.08 g	92.69 e	49.71 d	48.95 d
6-BA (20 ppm)	22.33 bcd	21.00 d	12.67 bcd	11.67 c	118.65 bcd	110.61d	66.38 bc	61.45 c
6-BA (40 ppm)	25.67 a	24.33 a	14.00 a	14.00 a	137.06 a	129.13 a	73.20 a	74.29 a
Ch (100 ppm)	20.00 ef	22 bcd	2.00 d	12.00 c	106.73 fg	116.9 bcd	64.04 c	63.76 c
Ch (200 ppm)	21.00 cde	22 bcd	13.33 abc	13.67 ab	112.14 def	116.68 bcd	71.21 ab	72.48 ab
TRIA (2.5 ppm)	22.67 bc	22.67 bc	12.33 cd	12.67 bc	121.11 bc	120.36 bc	65.90 bc	67.26 bc
TRIA (5 ppm)	23.67 b	23.33 ab	13.67 ab	13.33 ab	126.77 b	123.98 ab	72.22 ab	70.84 ab
KSi (100 ppm)	20.67 def	21.67 cd	12.00 d	12.67 bc	109.87 ef	115.19 cd	63.79 c	67.34 bc
KSi (200 ppm)	22.00 bcd	23 abc	13.67 ab	13.33 ab	117.99 cde	122.36 abc	75.09 a	70.93 ab

Means with different letters for each plant parameter are considered significantly different ($p<0.05$) using the Duncan's multiple range test.

Table 8 - Average of pod weight, pod length, fresh pods weight, and pods diameter of snap bean as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	Average of pod weight (g)		Pod length (cm)		Pods diameter (mm)	
	2021	2022	2021	2022	2021	2022
Control	5.27 b	5.25 c	12.97 b	13.27 cd	6.67d	7.67 b
6-BA (20 ppm)	5.31 ab	5.27 b	13.57 a	13.70 ab	8.00 abc	8.00 ab
6-BA (40 ppm)	5.34 a	5.31 a	13.70 a	13.77 a	8.33 ab	8.33 ab
Ch (100 ppm)	5.34 a	5.31 a	12.97 b	13.23 d	7.33 cd	8.33 ab
Ch (200 ppm)	5.34 a	5.30 a	13.10 b	13.30 cd	8.33 ab	8.67 a
TRIA (2.5 ppm)	5.34 a	5.31 a	13.40 ab	13.50 bc	8.00 abc	8.33 ab
TRIA (5 ppm)	5.36 a	5.31 a	13.57 a	13.67 ab	8.33 ab	8.33 ab
KSi (100 ppm)	5.32 ab	5.32 a	13.07 b	13.33 cd	7.67 ab	8.00 ab
KSi (200 ppm)	5.36 a	5.32 a	13.37 ab	13.47 bcd	8.67a	8.33 ab

Means with different letters for each plant parameter are considered significantly different (p<0.05) using the Duncan's multiple range test.

Table 9 - The contents of ascorbic acid, fiber, soluble sugar, and protein in snap bean pods as affected by 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), and potassium silicate (KSi) in the 2021 and 2022 seasons

Treatment	Ascorbic acid (mg 100 g FW^{-1})		Fiber (g 100 g FW^{-1})		Soluble sugar (g 100 g FW^{-1})		Protein (%)	
	2021	2022	2021	2022	2021	2022	2021	2022
Control	17.43 d	18.60 c	3.51a	3.49 a	2.13 d	2.19 c	17.94 f	18.25 e
6-BA (20 ppm)	18.53 cd	19.73 bc	3.36 c	3.35 d	2.21 c	2.19 c	18.69 de	19.31 c
6-BA (40 ppm)	19.57 abc	20.38 ab	3.31 d	3.31 e	2.25 b	2.23 b	19.75 ab	19.44 c
Ch (100 ppm)	19.70 abc	20.13 b	3.38 bc	3.36 d	2.20 c	2.21 bc	18.06 f	18.81 d
Ch (200 ppm)	20.70 a	21.30 a	3.39 bc	3.37 cd	2.21 c	2.21 bc	19.13 cd	19.44 c
TRIA (2.5 ppm)	19.31 bc	19.78 b	3.30 d	3.29 e	2.30 a	2.32 a	19.63 abc	19.88 b
TRIA (5 ppm)	20.44 ab	20.61 ab	3.28 d	3.28 e	2.33 a	2.32 a	20.00 a	20.31 a
KSi (100 ppm)	19.70 abc	20.07 b	3.40 bc	3.39 bc	2.20 c	2.21 bc	18.50 ef	18.81 d
KSi (200 ppm)	19.87 abc	20.07 b	3.42 b	3.41 b	2.21 bc	2.23 b	19.13 cd	19.63 bc

Means with different letters for each plant parameter are considered significantly different (p<0.05) using the Duncan's multiple range test.

sugar, and protein) of snap bean green pods were investigated. Except for plants treated with 6-BA at 20 ppm, the ascorbic acid content in the pods of treated plants was significantly increased with respect to the control in both cultivation seasons. Also, the treatments with 6-BA (40 ppm), TRIA (2.5 or 5 ppm), or KSi (200 ppm) significantly increased the amount of soluble sugar in the pods with respect to the control in both cultivation seasons. Moreover, it was observed that the application of 6-BA (20 and 40 ppm), Ch (200 ppm), TRIA (2.5 or 5 ppm), orKSi (200 ppm) resulted in enhancement of protein content within the pods, with respect to the control plants, across both seasons (Table 9).Comparable findings also demonstrated the beneficial effects of Cks, Ch, TRIA, orKSi on the levels of protein, soluble sugar, and ascorbic acid in cereal crops, vegetable, or legumes (Naeem *et al*., 2012; Artyszak, 2018; Hu *et al*., 2022). Moreover, snap beans should have fleshier green pods with little fiber content where immature pods are eaten as vegetables. In this study, the applied treatments significantly reduced the amount of fiber in the pods relative to the control group (Table 9). Overall, in both growth seasons, the TRIA (5 ppm) spraying treatment showed remarkable efficacy across all quality criteria.

4. Conclusions

The applications of 6-benzylaminopurine (6-BA), chitosan (Ch), triacontanol (TRIA), or potassium silicate (KSi) by foliar spraying have the potential to enhance the development and agronomic characteristics of snap bean plants.Specifically, the use of 5 ppm TRIA demonstrates the most advantageous improvements in growth, blooming, yield, and overall quality. This study could potentially establish a theoretical framework for improving the commercial production of snap beans in summer conditions. Also, by demonstrating the efficacy of biostimulants, sustainable agricultural practices can enhance food production and environmental stewardship in subtropical regions.

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