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Effects of two different doses of biofertilizer on the growth and production of Kohlrabi (*Brassica oleracea* var. *Gongylodes* L.)

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Abstract

Objective

This study was carried out to investigate the effects of two levels of biofertilizer on the growth and yield of cabbage (*Brassica oleracea* var. L.) under local environmental conditions under standard conditions involving water and sunlight exposure.

Materials and methods

A field experiment with biofertilizer (3,6 kg ha-1) was conducted in loam soil at a research field affiliated with a green gold company (Diyala City, Iraq). The physical and chemical characteristics of the field soil were recorded. The experimental methodology was a randomized complete block design (RCBD) with three replications versus the control. The first dose of biofertilizer (3), which consisted of *Trichoderma* and *Bacillus* strains with humic acids and seaweed extract, was applied one week after planting, and the second dose (6) was added after two weeks. In their plots, the Kohlrabi seedlings were manually planted in rows of two meters

each, separated by 20 cm (3 rows per plot). The seeds of Kohlrabi were sowed (on 10 September 2023 in the seedbed and planted in the field on 22 October 2023). Throughout the research, all weeding was performed by hand. The crop was harvested after 90 days of planting and statistically analyzed.

Results

The results indicated that the 6 kg/ha treatment resulted in the maximum average growth and productivity parameters, such as plant height, leaf number, chlorophyll content, leaf area, bulb length, bulb diameter, bulb weight, root weight and total yield. The values for these genes were 56.76 ± 2.30, 9.56 ± 0.23, 14.53 ± 0.26, 13.03 ± 0.61, 9.00 ± 0.05, 11.81 ± 0.13, 298.33 ± 4.41, 51.50 ± 0.50, and 9.54 ± 0.14, respectively, under the lowest values recorded under the control treatment.

Conclusions

This study is very significant because it provides an alternative for chemical fertilizers that ecologically enhances cabbage growth and production while lowering cost and keeping the soil and food healthy.

Keywords: biofertilizers, *Brassica oleracea* var. *Gongylodes*, Kohlrabi, plant growth promotion, soil fertility

Paper Type: Research Paper.

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Introduction

Kohlrabi (*Brassica oleracea* var. *Gongylodes* L.) is a member of the large, ecologically important cruciferous family and is known for its high nutritional value due to the widespread

presence of vitamins, minerals, fiber, and antioxidant dietary phenolic compounds (Abou-El-Hassan et al., 2020; Paśko et al., 2021). These properties make Kohlrabi a healthy dietary food and an economic source for farmers in many areas of production (Khatun et al., 2024). As the global interest in food security and sustainable agriculture continues to grow, enhancing crop productivity and quality is a major priority for scientists and producers. However, the indiscriminate use of chemical fertilizers subsequently increased crop productivity over the last few decades at the cost of serious environmental problems such as pollution of soil and water, long-term soil fertility declines and adverse impacts on human and animal health. This has resulted in a search for more sustainable and ecological options, especially biofertilizers. BOFs are microorganisms that have positive effects, such as nitrogen-fixing bacteria, phosphorus- and potassium-solubilizing bacteria, and symbiotic root fungi (mycorrhizae) (Nurhidayati et al., 2024). These factors all augment nutrient acquisition from the soil, improve the microbially balanced rhizosphere, and promote growth via the synthesis of natural plant growth regulators such as auxins and gibberellins (Saad et al., 2019, Altaai & Saad, 2024). Different studies have revealed that different biofertilizers are beneficial for improving vegetative growth, increasing biomass, increasing chlorophyll content, and increasing crop quality in other plants, such as tomatoes (Coulibaly et al., 2021), radishes (Naidu et al., 2023) and cabbage (Rana et al., 2020). Research on the effects of biofertilizers on Kohlrabi, especially the synergistic impact of biofertilizers on increasing the physiological and productive attributes of the crop, has not been conducted. The innovation of this study is the assessment of the interaction effects of various biofertilizers of interest on Kohlrabi growth and yield, which has not been significantly studied in contrast with other crucifer crops. This study also aims to associate the role of biofertilizers not only with productivity but also with the nutritional value and marketable quality of the crops, which is a major concern because consumers have now turned into healthy and safe food seekers. Accordingly, this research not only bridges a knowledge gap in the impact of biofertilizers on crops but also paves the way for developing environmentally friendly agricultural practices with sustainable economic returns. Thus, the present study aimed to describe the effects of two levels of biofertilizer on the growth and yield of Kohlrabi (*Brassica oleracea* L.) under various environmental conditions.

Materials and methods

Isolates used in cultivation: The isolates used in the study (*Trichoderma* and *Bacillus* strains with humic acids and seaweed extract) were obtained from the Directorate of Agriculture Research, Ministry of Science and Technology.

Field experiment: A field experiment with biofertilizer levels (3,6 kg ha-1) was conducted from 2023-2024 in loam soil in a research field affiliated with the green gold company located in Diyala Governorate. Table 1 displays a few measured physical and chemical characteristics of the field soil. This research contributes information regarding the impact of biofertilizer on the growth and production of Kohlrabi under various environmental conditions. The experimental methodology was a randomized complete block design (RCBD) with three replications. The first dose of biofertilizer (3), which consisted of *Trichoderma* and *Bacillus* strains with humic acids and seaweed extract, was applied one week after planting, and the second dose (6) was added after two weeks. In their plots, the Kohlrabi seedlings were manually planted in rows of two meters each, separated by 20 cm (3 rows per plot). The seeds of Kohlrabi were sowed (on 10 September 2023 in the seedbed and planted in the field on 22 October 2023). Throughout the research, all weeding was performed by hand. The crop was harvested after 90 days of planting and statistically analyzed.

Table 1. The field soil: its physical and chemical characteristics

Traits	Values
pH	7.8
EC, Ds/m-^{1}	2.21
Nitrogen, ppm	40
Phosphorous, ppm	12.5
Potassium, ppm	208
Sand, g.kg^{-1}soil	300
Silt, g.kg^{-1}soil	490
Clay, g.kg^{-1}soil	230
Soil texture	loam

Physiological measurements; Plant height (cm): Ten plants were chosen at random from each treatment, and their heights were measured from the base to the tip. The mean was then noted.

Leaf number per plant: For each treatment, the number of leaves was determined from 10 randomly selected plants.

Chlorophyll content (g/cm^2): Chlorophyll content was estimated for 10 samples of plant leaves at the flowering stage via Minolta SPAD 502 (Reynolds et al.,1998) via the following equation:

$$\text{Total chlorophyll} = \frac{\text{SPAD}*10.40-80.05}{10{,}000} = \text{g/cm}^2.$$

The leaf area (cm²): The total leaf area was calculated by adding the areas of all the leaves on each plant.

Bulb length (cm): Ten plants were chosen at random from each treatment, and their fruit lengths were measured.

Bulb diameter (cm): Fifty randomly chosen plants from each treatment had their fruit diameters measured.

Bulb weight (g): After the leaves were removed from the bulbs, the fresh weight was calculated.

Root weight (g): Three plants were dried in an oven at 70°C until the weight stabilized. The dry biomass was recorded and expressed in grams per plant.

Total yield: Fresh weights of leaves and bulbs (kg) from ten randomly chosen plants per plot were measured immediately after harvesting to compute the yield.

Statistical analysis: The study was carried out and examined as a factorial experiment with three replicates via a randomized complete block design (RCBD). Tukey's test was used to compare the mean values with a 5% chance.

Results

The results in Table 2 show that, compared with the control treatment, the biofertilizer treatments had a significant effect on all the studied traits of the Kohlrabi plants.

Plant height (cm): The results revealed that the greatest average plant height was achieved in the 6% treatment, with an average value of 56.76 cm, which was significantly greater than that in the control treatment (45.22 cm). The average height in the 3% treatment group was 51.12 cm, which was not significantly different from that in the control and 6% treatment groups. These findings indicate that the use of biofertilizers contributed to increasing the efficiency of nutrient absorption and stimulating vegetative growth through the secretion of natural growth regulators such as auxins and gibberellins, which positively impacted plant height.

Leaf number: Compared with the other treatments, the 3% treatment resulted in the greatest number of leaves (11.17 leaves/plant), which was clearly significantly different, whereas the control treatment resulted in the lowest number of leaves (8.85 leaves). In the 6% treatment, an average value of 9.56 leaves/plant was recorded between the two previous treatments. This is attributed to the ability of some of the enriched bacteria to improve vegetative growth by increasing the availability of nitrogen and phosphorus, which supports the formation of new leaves.

Chlorophyll Content: The results revealed that both treatments (3% and 6%) resulted in the highest chlorophyll content (13.20 and 14.53, respectively), which was significantly different

from that of the control treatment (10.13%). These findings indicate that biofertilizers enhance the efficiency of plant photosynthesis by increasing the absorption of major elements, especially nitrogen, which is a component of the chlorophyll molecule, thus increasing the photosynthetic efficiency and biological productivity of the plant.

Leaf area: Compared with the control, the treatments (3% and 6%) significantly outperformed the control, with average leaf areas reaching 12.36 and 13.03 cm², respectively, compared with 10.10 cm² in the control. This reflects the role of biofertilizers in increasing vegetative expansion, which contributes to the ability of plants to absorb light and increases their photosynthetic efficiency. P values revealed significant differences for all traits at the probability level ($P \leq 0.05$), most notably in the number of leaves ($P = 0.0003$) and chlorophyll content ($P = 0.0006$). The LSD test also confirmed the presence of clearly significant differences between treatments, with treatments 3 and 6 generally excelling in most of the traits studied.

Table 2. Effects of two levels of biofertilizer on plant height, leaf number, chlorophyll content and leaf area

Treatments	Plant height (cm)	Leaves number	Chlorophyll Content	Leaf area (cm²)
Control	45.22 ±2.69 b	8.85 ±0.12 c	10.13 ±0.13 b	10.10 ±0.45 b
Biofertilizers 3	51.12 ±2.38 ab	11.17 ±0.16 a	13.20 ±0.62 a	12.36 ±0.32 a
Biofertilizers 6	56.76 ±2.30 a	9.56 ±0.23 b	14.53 ±0.26 a	13.03 ±0.61 a
L.S.D.	8.549 *	0.625 **	1.377 **	1.637 **
P value	0.0446	0.0003	0.0006	0.010

*Different letters within the same column indicate significant differences at the probability level ($P \leq 0.05$).

**The presence of different letters in the same column indicates significant differences ($P \leq 0.01$).

The results in Table 3 show that, compared with the control treatment, the biofertilizer treatments had a significant effect on all the studied traits of the Kohlrabi plants.

Bulb Length (cm): An increase in the degree of biofertilizer led to an increase in the length of the bulb. Length was longer in the 6% treatment (9.00 cm) than in the control (6.61 cm) and 3% treatments (6.63 cm). This increase was due to the ability of the biofertilizers to promote vegetative growth and plant metabolism and the uptake of key nutrients such as nitrogen and

phosphorus. In addition, humic acid and seaweed extract promoted cell elongation and increased fruit size.

Bulb diameter (cm): Bulb diameter was significantly different only among the control (8.23 cm) and the 3% and 6% treatments (12.16 and 11.81 cm, respectively). This increase suggests that biofertilizers improved nutrient redistribution, cell adhesion, and fruit growth factors, contributing to an increase in the size and quality of the fruit.

Bulb weight (g): Compared with those in the control group (200 g), the heaviest bulbs in the 6% treatment (298.33 g) and 3% (240.35 g) treatment groups were significantly greater. These increases, which are in line with the increased length and diameter, indicate that the biofertilizers enhance carbohydrate and nutrient storage in storage tissues, increasing the final weight of fruits.

Root weight (g): A total of 51.50 g of root weight was recorded for the 6% treatment group, and 41.16 g of root weight for the control group increased significantly with increasing biofertilizer concentration. In addition to enhanced root health and functionality associated with more efficient absorption of nutrients and water, Trichoderma and Bacillus facilitate nutrient absorption over roots, which helps fruit to store more energy.

Total yield (kg/ha): The highest yield was recorded for the 6% treatment (9.54 kg/ha), followed by the 3% treatment (7.68 kg/ha) and the control (6.40 kg/ha). This significant increase reflects the complementary effect of biofertilizers with humic acid and seaweed extract in promoting plant growth, increasing storage tissue, and improving fruit quality. These results confirm the clear and synergistic effects of biofertilizers with organic components on fruit and root growth and total yield.

Table 3. Effects of two levels of biofertilizer on bulb length, bulb diameter, bulb weight, root weight and total yield

Treatments	Bulb length, cm	Bulb diameter, cm	Bulb weight, g	Root weight, g	Total yield kg ha $^{-1}$
control	6.61 ±0.19 b	8.23 ±0.14 b	200.00 ±0.10 c	41.16 ±0.60 c	6.40 ±0.03 c
Biofertilizers 3%	6.63 ±0.32 b	12.16 ±0.44 a	240.35 ±5.61b	45.55 ±0.06 b	7.68 ±0.18 b
Biofertilizers 6%	9.00 ±0.05 a	11.81 ±0.13 a	298.33 ±4.41 a	51.50 ±0.50 a	9.54 ±0.14 a
L.S.D.	0.746 **	0.966 **	14.260 **	1.565 **	0.464 **
P value	0.0003	0.0001	0.0001	0.0001	0.0001

** Values with different letters in the same column are significantly different (P≤0.01).

Figure 1. Effects of biofertilizers at the 6% level on Kohlrabi plants

Discussion

In addition to increasing fruit and root traits and total yield, biofertilizers and organic stimulants (*Trichoderma*, *Bacillus*, humic acid, and seaweed extract) had desirable effects on all vegetative and physiological parameters of Kohlrabi plants. Compared with the control, these treatments improved not only bulb length, diameter, weight, and root weight but also overall productivity. They also increased plant height, leaf number, chlorophyll content, and leaf area. These findings indicate that these components play important roles in enhancing the absorption of essential nutrients, particularly nitrogen, phosphorus and magnesium, as the increase in plant height, leaf area and chlorophyll content. The results revealed that, compared with noninoculated plants, biofertilizer-treated plants presented greater results in terms of the percentage of inoculated plants. Similarly, *Trichoderma* and *Bacillus* increase root growth while stimulating the secretion of plant growth regulators (auxins and cytokinins) (Satyal et al., 2024; Miljaković et al., 2020). Humic acid and seaweed extracts improve soil conditions, increase the solubility of nutrients and promote photosynthesis (Ismael and Sarhan, 2025; Ahmed, 2024). Similar results have been reported in other cruciferous crops, where PGPR and beneficial fungi increase vegetative growth and physiological characteristics under either particular or general conditions (Demir et al., 2023; Khan et al., 2009). The increase in traits such as bulb, root and yield in the organs in the treatments that included 3% and 6% biofertilizer also suggests an obvious synergetic effect of the factors involved. The highest bulb length, diameter and weight and the highest root weight and total productivity were observed in the 6% treatment group, which was attributed to the accumulation of carbohydrates and nutrients in storage organs and the increased uptake of nutrients and water by roots. Compared with the untreated control, seaweed extract stimulates cell growth by

promoting tissue elongation and expansion, as does humic acid, which increases the level of photosynthetic and nutrient uptake-related enzymes (Chen & Aviad, 1990; Terry et al., 2012). The leaf chlorophyll content is a growth parameter that increases with inoculation by biofertilizers. This increase in chlorophyll content is due to nitrogen fixation by biofertilizers, which increases the chlorophyll content in the leaves and then increases the efficiency of photosynthesis, which is reflected positively in the surface area of the leaves, and the highest total chlorophyll content is observed in dual compatible mixtures of inoculants. N is necessary in all vital processes that take place inside the plant, greatly affecting cell division, increasing meristem cell activity and thus expanding the surface area of the leaves (Kumari, 2017). Thus, the combined addition of beneficial microbes (*Trichoderma* and *Bacillus*) and organic stimulants (humic acid and seaweed extracts) consistently improved vegetative growth, physiological health, fruit size, root weight and kohlrabi productivity, combined with the effects observed with all individual additions. This finding indicates the possibility of employing this combination in integrated fertilization programs to decrease the dependence on chemical fertilizers and maximize yield and fruit quality without compromising sustainability.

Conclusions: The results revealed the growth, physiological, and productive traits of Kohlrabi (*Brassica oleracea* var. gongylodes). Compared with the control, the biofertilizer treatments resulted in greater height, leaf, chlorophyll and leaf areas, bulb length, bulb diameter, root weight, and productivity. The synergistic action of microbes and organic stimulants for better nutrient uptake, root stimulation and increased production of natural growth regulators might have contributed to this improvement. These results highlight the possibility of including these treatments along with integrated fertilization programs to improve productivity and quality while minimizing the use of chemical fertilizer.

Author contributions

Conceptualization: AFA and RJM; Data curation: DAS; Formal analysis: NMA; Methodology: NMJ; Project administration: AFA; Resources: DAS and NMA; Supervision: AFA and RJM; Validation: AFA and RJM; Visualization: AFA and RJM; Writing – original draft: AFA and RJM; Writing – review & editing: NMA.

Data availability statement

Available with the authors upon request.

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Ethical considerations

Approval was obtained from the University of Diyala, Iraq.

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Conflict of interest

No conflict of interest exists in the publication of this research**.**

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تأثیر دو دوز مختلف کود زیستی بر رشد و تولید کلم قمری (*Brassica oleracea* var. *Gongylodes* L.)

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چکیده

هدف: این پژوهش با هدف بررسی تأثیر دو سطح مختلف از کود زیستی بر رشد و عملکرد کلم قمری (*Brassica oleracea* var. L.) تحت شرایط محیطی محلی و در شرایط استاندارد شامل آبیاری و تابش نور خورشید انجام شد.

مواد و روش‌ها: آزمایش مزرعه‌ای با دو سطح کود زیستی (۳ و ۶ کیلوگرم در هکتار) در خاک لومی در مزرعه تحقیقاتی وابسته به شرکت "طلای سبز" در شهر دیاله (عراق) انجام شد. ویژگی‌های فیزیکی و شیمیایی خاک مزرعه ثبت گردید. طرح آزمایشی به‌صورت بلوک‌های کامل تصادفی (RCBD) با سه تکرار و یک تیمار شاهد اجرا شد. دوز اول کود زیستی (۳ کیلوگرم در هکتار) شامل سویه‌های Trichoderma و Bacillus همراه با اسیدهای هیومیک و عصاره جلبک دریایی، یک هفته پس از کاشت و دوز دوم (۶ کیلوگرم در هکتار) دو هفته بعد اعمال گردید. نشاءهای کلم قمری به‌صورت دستی در ردیف‌هایی به طول دو متر و فاصله ۲۰ سانتی‌متر (سه ردیف در هر کرت) کاشته شدند. بذرها در تاریخ ۱۰ سپتامبر ۲۰۲۳ در خزانه کشت و در تاریخ ۲۲ اکتبر ۲۰۲۳ به زمین

اصلی منتقل شدند. در طول دوره تحقیق، وجین به‌صورت دستی انجام شد. محصول پس از ۹۰ روز از کاشت برداشت و داده‌ها به‌صورت آماری تجزیه و تحلیل شدند.

نتایج: نتایج نشان داد که تیمار ۶ کیلوگرم در هکتار بیشترین میانگین رشد و شاخص‌های تولید را شامل ارتفاع گیاه، تعداد برگ، محتوای کلروفیل، سطح برگ، طول غده، قطر غده، وزن غده، وزن ریشه و عملکرد کل نشان داد. مقادیر این صفات به‌ترتیب برابر با ۲/۳۰ ± ۵۶/۷۶، ۰/۲۳ ± ۹/۵۶، ۰/۲۶ ± ۱۴/۵۳، ۰/۶۱ ± ۱۳/۰۳، ۰/۰۵ ± ۹/۰۰، ۰/۱۳ ± ۱۱/۸۱، ۴/۴۱ ± ۲۹۸/۳۳، ۰/۵۰ ± ۵۱/۵۰ و ۰/۱۴ ± ۹/۵۴ بود، در حالی که پایین‌ترین مقادیر مربوط به تیمار شاهد ثبت گردید.

نتیجه‌گیری: این پژوهش اهمیت بالایی دارد زیرا جایگزینی مناسب برای کودهای شیمیایی ارائه می‌دهد که رشد و تولید کلم قمری را به‌صورت زیست‌محیطی بهبود داده، هزینه‌ها را کاهش داده و سلامت خاک و محصول را حفظ می‌کند.

کلمات کلیدی: تحریک رشد گیاه، حاصلخیزی خاک، کلم قمری، کود زیستی، *Brassica oleracea* var. *Gongylodes*

نوع مقاله: پژوهشی

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