Yield and Fruits Quality of Potted Strawberry Plants Grown with Bacillus-Coated NPK Fertilizer

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ABSTRACT

NPK fertilizer coated with rhizobacteria biofertilizer Bacillus supports biofertilizer use while reducing the amount of NPK fertilizer needed. The aim of the greenhouse experiment was to analyze the growth and fruit production of potted strawberries after applying Bacillus-coated NPK 16-16-16 (BCN); and BCN's potency to reduce NPK doses. The study was arranged as a Completely Randomized Block Design testing four different combinations of BCN dosage and type. The control treatment involved no fertilizer and a complete and a half dose of NPK fertilizer 16-16-16. The results showed that the shoot height and the fruit number of the strawberry cv Festival were not affected by BCN application but fertilizer type significantly influenced fruit weight and sweetness. A full dose of BCN formula C produced the most significant fruit weight, but the highest sweetness came from plants treated with half the dose of NPK conventional fertilizer. However, one doses of BCN formula G reduced the fruit sweetness. This experiment shows the potential of BCN to support strawberry production in a greenhouse, but a longer duration of the experiment is needed to obtain more harvest data.

Keywords: Biofertilizer, Greenhouse cultivation, Plant Nutrition, Sweetness

INTRODUCTION

Strawberry fruit is an essential commodity in West Java, especially for Bandung, West Bandung, Cianjur, and Garut Regency. West Java supplies about 60% of strawberries to the national market. Strawberry cultivation is taken place in the highlands and mountainous areas because it requires low temperatures of 20-24^{0}C for the best growth (Asadpoor and Travallali, 2015).

In addition to temperature, fertilization is vital for promoting the growth and fruiting of strawberries (Khalil and Agah, 2018). Today, strawberries are grown in soilless substrates that depend on nutrient supply from liquid or solid inorganic fertilizers. The cost of chemicals used as raw materials for fertilizer production is currently rising. Therefore, the use of chemical fertilizers should be decreased without losing essential nutrients. Biofertilizers are widely proposed as an effective means to mitigate the reliance on chemical fertilizers. Specific soil-beneficial bacteria can supply critical nutrients, especially N, P, and K, along with growth factors that enhance plant development (Hayat *et al*., 2010; Setiawati *et al*., 2023).

The use of biofertilizers in strawberry plantations has been reported elsewhere. PGPR-based inoculants enable strawberry plants to mitigate photochemical limitations caused by abiotic stress, including drought (García-Lopez *et al*., 2024). A combination of biofertilizer and vermicompost is reported to improve plant growth, yield, and fruit quality in strawberries (Negi *et al*., 2021). The combined application of humic substances, biofertilizer, and NPK fertilizer increased total nitrogen and phosphorus, as well as the population of nitrogen-fixing bacteria in the growth substrate (Hindersah *et al*., 2023a). The challenge of using biofertilizers arises when the crop production costs increase due to the separate application of biofertilizers from NPK fertilizers. The Faculty of Agriculture, Unpad, in collaboration with PT Pupuk Kujang, developed a Bacillus-coated NPK 16:16:16 (BCN). Many Bacilli are rhizobacteria with inherent phosphate-solubilizing properties that benefit plants by making phosphate available. These soil-beneficial microbes are also known for their ability to produce growth factors, particularly phytohormones such as auxin, cytokinin, and gibberellin, to facilitate plant growth (Prakash and Arora, 2019; Ambawade and Pathade, 2015; Baghaee-Ravari and Heidarzadeh, 2014).

Research on microbial-coated NPK fertilizers remains limited. In lettuce plants, BCN maintains the uptake of N, P, and K, as well as lettuce yield, compared to NPK 16:16:16 application (Hindersah *et al*., 2023b). The study involving bacterial-coated NPK has not yet been reported for strawberries. However, a previous study showed that Azotobacter-Bacillus-coated Urea could reduce the use of NPK fertilizer in strawberry nurseries (Hindersah *et al*., 2021). We hypothesized that BCN could replace or reduce the dose of conventional NPK without compromising strawberry yield or quality. This pot experiment was conducted to observe plant growth, fruit yield, and the sweetness of strawberry fruit grown in the greenhouse after applying two doses and two types of Bacillus-coated NPK 16:16:16; as well as to analyze the potency of BCN to reduce NPK doses.

MATERIAL AND METHODS

The Soil Biology Laboratory, Faculty

of Agriculture, Universitas Padjadjaran, provided the Bacillus-coated NPK formulas C and G. In the balanced population, the Bacillus consortium comprised *B. safeness* B1, *B. subtilis* D2, *B. altitudinis* B14, and *Bacillus* sp. E2. The experiment was conducted in CV Bumi Agro Technologi, Bandung Barat Regency, from September to December 2021.

Experimental Design

A pot experiment was conducted in the greenhouse, which was set up in a randomized block design with four treatments and five replications. Therefore, there were 20 experimental pot units. The treatments were four combinations of dosage (complete and half of the dosage) and type of BCN, including formula C (BCN-C) and formula G (BCN-G). Three control treatments were used, including one without fertilizer and one with one-half of the dosage of NPK 16-16-16. The complete dosage of both fertilizers follows the recommended dosage for strawberries, 200 kg ha^{-1}. The amount of complete fertilizer dosage was calculated based on the NPK recommended dosage and plant density in a hectare (100,000 plants). Each pot with one and a half dosages of BCN and NPK received 2 g and 1 g of fertilizer per plant, respectively. Two plants were grown in a pot and received equal fertilizer.

Experimental Establishment

The strawberry cv Festival was planted in a substrate made of cocopeat, chicken manure, and sheep manure with a volume ratio of 8:2:1. A total of 10 kg of planting media was placed into a 40x40 cm polyethylene bag and incubated in a greenhouse for two days. Each pot was planted with two 6-week-old strawberry seedlings.

According to the dosage, Bacillus-coated NPK and conventional NPK (16-16-16) were added to the soil three and 30 days after planting. The fertilizer was applied about 4-5 cm from the stem (Figure 1) and immediately covered with substrate. All plants were kept in a greenhouse for 6 weeks.

Figure 1. Fertilizer placement near the plant stem.

The average height of two plants in a pot was measured 1-4 weeks after the initial BCN treatment. Strawberries were harvested twice a week starting from the first fruiting (2 weeks after the initial BCN application) and continued until 4 weeks later. Harvest fruits were characterized by 80% of red color. Yield parameters included the weight of individual fruits and the number of strawberries harvested each week from each plant in a pot. The average sweetness content of strawberries harvested from the two plants was measured using a Brix meter. Figure 2 shows the timeline from planting to harvesting.

Analysis of variance (ANOVA) was conducted for all plant and fruit parameters at $p<0.05$. Duncan's Multiple Range Test (DMRT) was done for the parameters significantly affected by treatments based on ANOVA. The growth data were presented with standard deviation. However, the yield data were presented without standard deviation due to the asynchronous maturation of strawberry fruits among the plants.

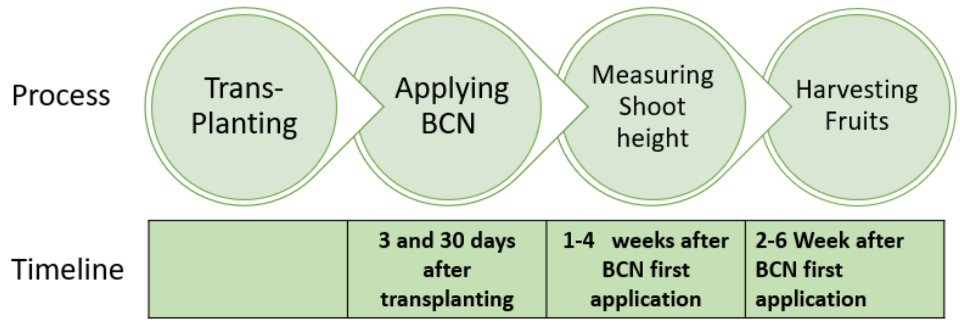

Figure 2. Timeline of experimental establishment on the use of BCN for potted strawberries

RESULTS AND DISCUSSION

Plant Height

The ANOVA verified that the NPK fertilizer treatments did not influence the shoot height. Table 1 indicates that, regardless of the fertilizer treatment, the average shoot height of strawberries was about 21-22 cm. The shoot height of plants with any dose and type of BCN did not significantly differ from that of plants without fertilizer or those given NPK conventional fertilizer. The strawberry plants grew well in the cocopeat-based soilless medium; the number of leaves on each plant was maintained at 5-6 by removing old and damaged leaves. The shoot height of the plants remained steady until 4 weeks after transplanting, even though fertilizer was applied 3 weeks after transplanting.

Table 1. Shoot the height of the strawberry following the application of Bacillus-coated NPK

Fertilizer Treatments	Shoot height (cm) at			
	week* 1	Week 2	Week 3	week 4
Control	22.6 ± 1.52	21.8 ± 1.97	21.8 ± 1.97	22.0 ± 1.70
1 dosage of NPK	21.8 ± 2.68	20.1 ± 2.14	20.2 ± 2.74	21.4 ± 1.02
½ dosage of NPK	22.9 ± 2.30	20.6 ± 2.48	20.8 ± 2.66	21.4 ± 1.18
1 dosage of BCN-C	22.9 ± 3.01	21.6 ± 1.08	22.1 ± 1.29	21.6 ± 1.08
½ dosage of BCN-C	24.6 ± 1.82	21.8 ± 3.47	21.3 ± 3.15	22.2 ± 3.09
1 dosage of BCN-G	22.8 ± 1.35	20.9 ± 2.30	20.9 ± 2.30	21.3 ± 1.79
½ dosage of BCN-G	22.4 ± 0.65	20.7 ± 2.31	20.7 ± 2.31	21.3 ± 1.25

*Week after the first NPK and BCN application

Strawberry Yield

Harvesting began two weeks after the first BCN application (Figure 3). Fruit harvesting was not performed at the same time for each treatment and replication because the anthesis stage of the strawberry plants varies from one plant to another. The anthesis phase started three to four days after the initial fertilizer application, which was three days after planting. The average number of fruits per plant was calculated from five replications of each treatment.

The experiment found that fertilizer treatment did not affect the number of fruits. However, at the 3rd harvest, plants treated with half the dosage of

BCN-G had significantly higher fruit counts (Table 2). The fruit numbers of plants receiving half the dose of NPK conventional were similar to those treated with a full dose of BCN-G; both were 20% lower than the ½ dose of BCN-G. At the 4th and 5th weeks of harvest, each plant from different treatments produced a single fruit (Table 2) during each harvest.

Table 2. Number of fruits per potted strawberry after application of Bacillus-coated NPK

Fertilizer Treatments	The number of fruits harvested at				
	week* 2	week 3	weeks 4	week 5	week 6
Without fertilizer	0.8	0.4	0.4 ab	1.0	1.0
1 dosage of NPK	1.0	0.2	0.4 ab	1.0	1.0
½ dosage of NPK	0.8	0.8	0.8 bc	1.0	1.0
1 dosage of BCN-C	1.4	0.4	0.2 a	1.0	1.0
½ dosage of BCN-C	0.6	0.2	0.2 a	1.0	1.0
1 dosage of BCN-G	1.0	1.2	0.8 bc	1.0	1.0
½ dosage of BCN-G	1.0	1.0	1.0 c	1.0	1.0

Numbers followed by the same letter in each column do not show significant differences according to Duncan's Multiple Range Test at $p \leq 0.05$. *Week after the first NPK and BCN application.

Table 3 displays a significant difference in fruit weight among plants treated with different fertilizers from the 2nd to the 3rd harvest week. The fruit weight varied based on the treatment and harvest time. Plants that received no fertilizer had the lowest fruit weight at each harvest. Fruit weight was higher during the fourth harvest compared to other harvests, although weekly data did not show statistical significance. Plants treated with one and a half dose of BCN-G consistently produced the heaviest fruit compared to plants without fertilizer and those treated with NPK or BCN-G.

Table 3. Fruit weight of strawberry after application of Bacillus-coated NPK

Fertilizer Treatments	Fruit weight (g) per plant harvested at				
	week*2	week 3	weeks 4	week 5	week 6
Without fertilizer	5.07	2.24 a	2.68 b	5.62 a	5.96 a
1 dosage of NPK	8.01	1.98 a	3.71 b	7.24 a	8.31 b
½ dosage of NPK	6.86	10.28 b	4.5 ab	8.54 ab	8.35 ab
1 dosage of BCN-C	8.41	7.21 ab	1.0 a	10.33 b	8.51 b
½ dosage of BCN-C	6.75	2.05 a	1.56 ab	10.91 b	8.16 ab
1 dosage of BCN-G	9.17	6.41 ab	5.17 bc	14.47 c	7.12 ab
½ dosage of BCN-G	12.30	6.08 ab	6.79 c	15.87 c	8.34 ab

Numbers followed by the same letter in each column do not show significant differences according to Duncan's Multiple Range Test at $p \leq 0.05$. *Weeks after the first NPK and BCN application.

According to the consensus, the Brix scale measures total dissolved solids (mainly sugar) in a liquid. One degree Brix equals one gram of sucrose in 100 grams of solution. The Brix degree decreased from the first week to the third week of harvest, then increased up to the fifth week of harvest (Table 4). At the fifth week of harvest, the sweetness of fruit with half the dosage of conventional NPK was 8.5, the highest among the Brix readings of fruits with other fertilization treatments.

Table 4. Brix scale (°Bx) of strawberry fruit harvested from the plants treated with Bacillus-coated NPK and NPK fertilizer

Fertilizer Treatments	Brix Degree of the Strawberry Fruit Harvested at				
	week* 2	week 3	weeks 4	week 5	week 6
Without fertilizer	3.74	1.05	2.80 ab	7.18	7.00 bc
1 dosage of NPK	5.66	1.40	2.60 ab	7.72	7.40 bc
½ dosage of NPK	4.94	5.68	6.06 bc	7.20	8.5 c
1 dosage of BCN-C	5.39	4.36	0.00 a	7.02	7.00 bc
½ dosage of BCN-C	5.52	1.50	2.02 ab	7.28	7.40 bc
1 dosage of BCN-G	6.90	4.91	6.52 bc	7.40	4.40 a
½ dosage of BCN-G	8.72	4.15	8.84 c	8.92	6.40 b

Numbers followed by the same letter in each column do not show significant differences according to Duncan's Multiple Range Test at $p \leq 0.05$. *Weeks after the first NPK and BCN application; **absence of ripe fruits.

Strawberry seedlings (daughter plants) were grown from runners of mother plants. The growth and yield of strawberries are heavily influenced by the quality of these seedlings, which depends on the health of the mother plants. Physiological uniformity among daughter plants ensured similar growth until the second fertilization, occurring three weeks after transplanting. Plant height was only measured up to the fourth week after planting (see Table 1). One month of measuring plant height confirmed that the BCN provided similar available nutrients to NPK, as the Bacillus in the BCN had not yet played an essential role in plant growth. The results align with studies on lettuce and tomatoes, which showed that any dosage of BCN and NPK did not impact plant growth. (Hindesah *et al*., 2023b; Hindersah *et al*., 2024).

The anthesis phase is not simultaneous, leading to different fruiting times and maturity levels for each plant. Harvesting over five weeks explains the increase in fruit number per plant (Table 2), but likely decreases the fruit weight. The first harvest yields fruits with higher weight than later harvests. The initial fruit comes from the first flower in each inflorescence, with only a few ripe fruits. In later harvests, more than one flower and fruit are present, so photosynthates are divided among many fruits, making them smaller and lighter. However, at the fifth harvest, plants treated with a full dose of BCN-C had the highest fruit weight. Meanwhile, plants given half the NPK dosage produced fruits with higher sweetness, comparable to those with one and a half times the BCN-C dose. Surprisingly, applying BCN-G significantly reduced fruit sweetness.

The Bacillus bacteria improved the quality of the strawberries rather than increasing their quantity. The sweetness of strawberries is mainly due to the accumulation of sugars like sucrose, fructose, and glucose as the fruit ripens (Taghavi *et al*., 2019), which are essential metabolites produced by photosynthesis. Although it does not affect plant growth, Bacillus might increase the sugar content in plants as a result of photosynthesis that correlates with the N in the chloroplast. The N content of strawberry shoots was not measured; however, the ability of Bacillus to fix N has been reported (Jain *et al*., 2021). Therefore, Bacillus could contribute to N availability in the soil. The sweetness is also influenced by the phytohormone auxin (Guo *et al*., 2022).

All Bacillus species coated onto the surface of BCN produced auxin in vitro, which can enhance the sweetness of the fruit. However, none of the Bacilli were identified to fix the dinitrogen, so in this experiment, the nitrogen availability for the plant relies on the NPK fertilizer. The BCN-G contained higher enrichment minerals than BCN-C, which reduced the NPK weight of BCN-G compared to BCN-C. Therefore, N supply BCN-G might not be sufficient for glucose synthesis. Nonetheless, further comprehensive research is needed to obtain a detailed correlation between the nitrogen content in BCN and the sweetness of fruit.

The highest °Brix value of fruits is 8.5 with the average of 5.6. The Strawberry cv. Festival was introduced from the subtropical region of USA and became a “local cultivar”. Growing strawberry in tropical areas face a significant challenge of high temperatures that reduce the sweetness. The Brix degree of Strawberry cv. Festival in subtropical is between 8.44 and 10.51 (Murthy *et al*., 2017)

CONCLUSION

The BCNs can replace NPK fertilizer up to 50% to produce a similar strawberry yield, even though the plant growth was not changed. The shoot height of the potted strawberry stayed identical for up to 4 weeks after applying NPK and BCN. The sweetness of the strawberry fruit was inconsistent at all times of harvest. However, the same Brix value was obtained from fruits treated with all fertilizers. The experiment verified that BCN can be introduced in strawberry cultivation in the greenhouse. Nonetheless, Longer-term research is needed to observe the effects of BCN on fruit productivity and sweetness, as well as other fruit qualities such as fruit dimension and sugar and acid content.

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