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Increased Production and Flavonoids of Two Celery Highland Varieties (*Apium graveolens* L.) by Endophytic Bacteria in Lowland

Andika Akbarul Iman, Sandra Arifin Aziz*) and Abdul Munif

IPB University, Bogor, West Java, Indonesia

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*) Corresponding author:
E-mail: *) sandraaziz@apps.ipb.ac.id

ABSTRACT

Celery (*Apium graveolens* L.) is an annual plant that grows optimally in Indonesia's highlands. The provision of endophytic bacteria has the role of plant protection, increasing agronomic growth and plant physiology, and overcoming environmental stresses. This study aims to assist the adaptation of highland celery varieties in the lowlands due to the decreasing agricultural highland and increase their production with endophytic bacteria applications. The experiment was executed in Bogor, Indonesia using a randomized block design with two factors. The celery variety served as the first factor [Amigo and Aroma (highland variety), and Summer Green (lowland variety)], and endophytic bacteria was the second factor (control, APE35, a combination of APE35+BAT, APE35+BAT+EQ26, and the PTM3 consortium). The result showed that endophytic bacteria helped the plants to adjust growth from highland to lowlands and increase yield. In lowland, Amigo showed shorter in height but it has a significantly higher stalk number, carotenoids, and flavonoids than Summer Green (lowland control). Endophytic bacteria increased plant growth, N, P, pigments, and flavonoids. APE35 or PTM3 with Amigo produced significantly higher leaf area, number of tillers, number of leaflets, leaf area, head fresh weight (78.529 and 75.054 g/plant), total fresh weight (81.67 and 85.395 g/plant), and total flavonoids (1484.818 and 1502.459 mg QE/100 g) than Summer Green without endophytic bacteriar.

INTRODUCTION

Celery (*Apium graveolens* L.) is a plant that has benefits as a medicinal plant (Handayani & Widowati, 2020). It has antifungal activity, lowers blood pressure and cholesterol, healthy joints, anti-inflammatory, antiseptic properties, diuretic, smooth nerves, relieves pain (Fazal & Singla, 2012), antihypertensive (Kumar & Pandey, 2013), and anticancer (Arisandi & Sukohar, 2016). This is because celery contains secondary compounds or secondary metabolites like flavonoids that act as antioxidants that protect the body and cells from damage caused by free radicals (Kumar & Pandey, 2013). The origin of celery is from the subtropical area. Indonesia is a tropical country with limited arable highlands. Celery is one of the subtropical-originated plants that is widely grown in Indonesia in highland areas (>700 m.a.s.l.) with a temperature of 15-24°C. The popular celery type in Indonesia is the leaf celery or *Apium graveolens* var. secalinum used mostly as herbs or vegetables (Lakitan et al., 2021).

Agricultural development in Indonesia has closely followed population growth and its geographical distribution was mostly in cities. Land use in Indonesia consisted of arable land 12.97%, permanent crops 12.14%, and other 74.88% (FAO ORG, 2022). The area of the highlands in Indonesia is small and mainly used for tourism purposes for the people from the cities. The limited arable land for vegetable production highland has made

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cultivation in lowland an opportunity to be explored. The highland celery varieties can be grown in lowland tropical climates but under stress. Providing shade will help the plant withstand the intensity of sunlight and high humidity. Celery, however, are less resistant to rain, with optimum rainfall ranging from 60-100 mm/month (Jannah, 2016).

Microorganisms such as endophytic bacteria and less optimal plant growing environment can increase the content of secondary metabolites (Kumar et al., 2021). Endophytic bacteria grow and colonize plant tissue and improve plant growth. It can produce growth-promoting substances, fix nitrogen, mobilize phosphate, and play a role in plant health. Endophytic bacteria have many abilities, including producing the hormone indole acetic acid (IAA), which plays an active role in the growth process of plant cells (Herlina et al., 2017). Endophytic bacteria can reduce damage and weather resistance and help plants tolerate the environment's extreme stress on biotic and abiotic factors (Trivedi et al., 2017). The endophytic bacteria can also protect host plants from competition from other bacteria (Afzal et al., 2019). It provides 60-80% of the N through its ability to fix nitrogen from the air thus making it suitable as a biofertilizer (Yulianti, 2012). This endophytic bacteria's ability increases plant products such as the fresh weight of stems, leaves, and shoot (Gusmaini et al., 2013). In addition, endophytic bacteria can also increase the production of natural bioactive compounds such as secondary metabolites (Bano et al., 2016).

Secondary metabolites are natural substances produced but do not directly affect growth and development, i.e., alkaloids, flavonoids, saponins, tannins, steroids, and triterpenoids. Flavonoids are found in almost all types of plants. Flavonoids are an important class of natural products. In nature, flavonoid compounds are products extracted from plants, and they are found in several parts of the plant. Flavonoids are used by vegetables for their growth and defense against plaques. Flavonoids protect plants from different biotic and abiotic stresses (Okungbowa et al., 2019) and act as unique UV filters, function as signal molecules, allopathic compounds, phytoalexins, detoxifying agents, and antimicrobial defensive compounds. Flavonoids have roles against frost hardiness, and drought resistance and may play a functional role in plant heat acclimatization and freezing tolerance (Panche et al., 2016). It can induce the formation of certain compounds as a response to plant defence, protecting against environmental stress, protecting against pests and diseases (phytoalexin), protecting from ultraviolet light, as a growth regulator, and competing with other plants (alelopathy) (Angin et al., 2019). Production of secondary metabolites will increase in the presence of stress on plants' environmental conditions. It is used as a strategy to increase the production of secondary metabolites in plants, and plant responses will reduce the productivity of primary metabolites and increase secondary metabolites as a defence against the environment (Rudin, 2020). Planting celery in the lowlands causes stress, producing higher flavonoids, and administering endophytic bacteria would be one solution. This study aims to evaluate the role of endophytic bacteria in highland celery (*Apium graveolens* L.) adaptation in lowlands by producing higher flavonoids and increasing its productivity through IAA production, converting urea into ammonium, dissolving phosphate, and increasing growth and production variables.

MATERIALS AND METHODS

Celery Varieties and Endophytic Bacterial Isolates

The research used celery varieties of Amigo, Aroma, and Summer Green types. The endophytic bacterial isolates used were APE35 single isolate from ferns, BAT from mangroves, EQ26 from oil palm, and PTM3 consortium isolates from mimosa. The endophytic bacterial isolates were collected from the Department of Plant Protection, IPB University, Bogor. The research was conducted at the Teaching Farm Sawah Baru Experimental Station of IPB University Bogor in January-May 2021.

Endophytic Bacterial Suspension Rejuvenation and Preparation

The endophytic bacterial isolates were rejuvenated on the streak plate (Walpajri et al., 2014) using TSA media (Tryptic Soy Agar) and incubated. The density test of endophytic bacterial isolates suspension was conducted based on a duplicate method. One ml suspension was transferred using a sterile micropipette into a test tube containing 9 ml of pure distilled water (to obtain a 10^{-1} dilution). The one ml suspension from a 10^{-1} dilution was transferred with a micropipette into a 10^{-2} tube until the dilution was 10^{-8}. Then, 0.1 ml suspension is taken from each dilution and put into a petri dish containing solid TSA media, then incubated at room temperature.

Immersion (Seed) Treatment

Seed celery soaked in a suspension of endophytic bacteria according to the treatment for 60 minutes (Nasib et al., 2016). Sterilized media containing the mixture of rice husk, top soil and cocopeat was used for seed germination. Nursery preparation is carried out on vertical shelves or seedling trays, and nurseries are carried out in rows (one row per shelf). Celery began to germinate 14 days after planting (DAP) with 80% Amigo germination, 85% Aroma, and 78% Summer Green. Celery seedlings were transferred to polybags after the plants had 3-4 fully expanded leaves at 46 DAP.

Implementation in the Field

This experiment was carried out in Bogor, Indonesia using a Completely Randomized Block Design with two factors. The first factor was celery varieties namely Amigo (highlands), Aroma (highlands), and Summer Green (lowlands). The second factor was endophytic bacterial isolates: no endophytes (control), single isolate APE35, two combinations of APE35 and BAT, three combinations of APE35, BAT, and EQ26, and isolates from the PTM3 consortium. There were 15 treatment combinations and four replications with 60 experimental units. Polybag used was those with the size of 30 cm x 30 cm containing 4.5 kg media which had been sterilized. The polybag was placed with the distance of 20 cm x 20 cm (Hendrika et al., 2017). Watering was carried out according to field capacity (Haridjaja *et al*. 2013). The endophytic bacterial suspensions were applied three times, i.e 10 ml at 2 WAP, 15 ml at 4 WAP and 20 ml at 6 WAP. Fertilization was applied between 2 and 6 weeks after planting (WAP) using NPK fertilizer (16:16:16) based on recommendation dosage. Control of pests and diseases applied when necessary. The applied pesticides were profenofos, amistar, difeconazole and azoxystrobin at recomemnded dosage. The harvesting was 90-100 DAP depending on varieties.

Physiological Analysis

NPK concentration analysis was carried out by collecting 8-10 g of dry powder simplicia of each sample treatment and the test were conducted based on the Kjeldahl method, titrimetric, and UV-VIS spectrophotometer. Analysis of chemical compounds anthocyanins, carotenes, and total chlorophyll, content these compounds in plants based on Sims and Gamon (2002). The bioactive analysis to find the content of total flavonoid was carried out according to Pothitirat et al. (2009). The analysis process is carried out at the Laboratory of the Research Centre Postharvest Development, IPB University.

Data Analysis

Data analysis was carried out on agronomic and physiological parameters. These parameters were plant height, head diameter, number of stalks, number of tillers, leaf area, number of leaflets, the root fresh weight, head diameter, head dry weight, head fresh weight, entire plant fresh weight, carotene, anthocyanin, total chlorophyll, total flavonoid, and plant NPK. The data obtained were analyzed using ANOVA (analysis of variance) at a 95% confidence level. The significantly different variables were furtherly tested using HSD test using R studio software.

RESULTS AND DISCUSSION

Bogor Climatology Station data during the study showed an average rainfall of 564.18 mm/month, an average maximum temperature of 31.75°C, an average minimum temperature of 22.35°C, and an average humidity level of 76.99 %. Observational data showed that the variety significantly affected plant height, number of stalks, and total flavonoid concentration. Endophytic bacteria significantly affected plant height, number of stalks, crown diameter, N and P nutrient concentrations, root fresh weight, and total flavonoids contents. The interaction of varieties with endophytic bacteria significantly affected the number of leaflets, number of tillers, leaf area, total chlorophyll, and total fresh weight of plants at the end of the experiment.

Physiological Test of Endophytic Bacteria Isolate

Physiological test of endophytic bacterial isolates related to indole acetic acid (IAA) based on Salkowsky reagent and L-Tryptophan as precursors of IAA biosynthesis resulted in an apparent color change to pink. All endophytic administrations produced IAA. The role of endophytic bacteria in promoting celery plant growth is because it stimulates the hormone IAA or auxin, which functions for plant growth and development. Plants need the IAA hormone or auxin produced by endophytic bacteria at specific concentrations totals. It plays a role in cell division in meristematic areas to stimulate plant growth (Kartikawati & Gusmaini, 2018).

Urease tests show the ability of bacteria to convert urea into ammonium to promote growth. The results showed that a change indicated media and positive colors from yellow to pink (Ulfa et al., 2016). All isolates of endophytic bacteria tested showed positive results.

In phosphate solvent testing using Politkovskaya's agar media with a mixture of phosphate content in the media, the results of the phosphate solvent test were seen as clear zones around the endophytic bacterial isolates on Politkovskaya's agar media (Sarker et al., 2014). Four isolates were tested, a single isolate APE35 and an isolate combination capable of dissolving phosphate marked with a clear zone. PTM3 endophytic bacteria isolate was not able to dissolve phosphate. The catalase test was used to determine the activity of bacteria to break down H_2O_2 into H_2O (water) and O_2 (oxygen). The appearance of froth or bubbles produced from all endophytic isolates showed that the bacteria have catalase activity.

Celery Varieties or Endophytic Bacteria on Agronomic Variables

The height of the Summer Green plant was significantly higher than Aroma and Amigo at 2 to 8 WAP. The Summer Green variety is a lowland variety that can grow and adapt optimally to the growing environment in the lowlands. The treatment of PTM3 endophytic bacteria was significantly higher than the control. All varieties treated with endophytic bacteria showed taller plants compared to those without bacteria (Table 1). The number of leafstalks in the Amigo is significantly higher than in the Aroma variety and did not differ from the Summer Green. Amigo experienced an increase in number of leafstalks because PTM3 gave significantly higher yields from 4 to 8 WAP than controls but did not differ from the other endophytic treatments (Table 2).

Table 1. Plant height of celery varieties under different endophytic bacteria treatments

Varieties	Plant height (cm) (WAP)			
	2	4	6	8
Amigo	11.89^{b}	19.68^{c}	30.49^{b}	32.95^{b}
Aroma	12.88^{b}	21.45^{b}	30.92^{b}	33.76^{ab}
S. Green	14.923^{a}	24.73^{a}	33.86^{a}	35.39^{a}
Endophyte	**Plant height (cm) (WAP)**			
	2	**4**	**6**	**8**
Control	13.06^{ab}	19.51	28.68^{c}	31.42^{b}
APE35	13.93^{a}	23.20^{a}	33.05^{ab}	34.42^{ab}
PTM3	14.01^{a}	23.14^{a}	35.52^{a}	36.82^{a}
APE35 and BAT	13.37^{ab}	22.75^{a}	30.55^{bc}	33.22^{ab}
APE35, BAT, and EQ26	11.79^{b}	21.17^{ab}	31.00^{bc}	34.29^{ab}

Remarks: The numbers followed by the same letter in the same column are not significantly different based on 5% HSD test; WAP = week after planting

Table 2. Number of stallks of celery varieties under different endophytic bacteria treatments

Varieties	Number of stalks (WAP)			
	2	4	6	8
Amigo	4.3^{a}	9.9^{a}	13.5^{a}	222^{a}
Aroma	4.3^{a}	8.2^{b}	12.2^{b}	20.8^{b}
S. Green	4.4^{a}	9.2^{ab}	12.9^{ab}	21.0^{ab}

Endophytes	Number of stalks (WAP)		
	4	6	8
Control	7.5^{b}	11.2^{b}	19.8^{c}
APE35	10.01	13.41^{a}	22.5^{ab}
PTM3	9.9^{a}	14.0^{a}	23.3^{a}
APE35 and BAT	9.0^{ab}	12.7^{a}	20.5^{c}
APE35, BAT, and EQ26	9.0^{ab}	12.8^{a}	20.6^{bc}

Remarks: The numbers followed by the same letter in the same column are not significantly different from the 5% HSD test; WAP = week after planting

The crown diameter with PTM3 endophytic bacteria was significantly higher than the control and not different with APE35 and three combinations of APE35, BAT, and EQ26. Root fresh weight was markedly higher in PTM3 endophytes than in control but not different with APE35. APE35 and BAT and three combinations of APE35, BAT, and EQ26 were the same as controls. The head dry weight reflects the plant biomass produced through photosynthesis. The results showed that the application of APE35 and PTM3 significantly increased head dry weight compared to controls. The combination of endophytic bacterial isolates was not different from the control. The possibility is that combining three types of endophytes has a commensalism effect on celery. The application of APE35 was significantly higher than the control and was not different from the endophytic bacterial isolates of PTM3 and the two combinations of APE35 and BAT (Table 3).

Endophytic bacteria can increase plant growth and development, such as plant height, number of stalks, and crown diameter. Endophytic bacteria penetrate plant roots by entering various organs. It also enters through wounds, intracellular spaces, or enzymes. Soaking and moistening the soil with the solution can promote plant growth and development effectively. In addition, endophytic bacteria are mutualistic and interact with the host plant. It can effectively increase the root fresh weight and improve the crown weight and total plant weight, and increase the productivity of celery plants. Endophytic bacteria can increase nitrogen fixation, photosynthetic activity, and indole acetic acid (IAA) production. The bacteria bind N_2 to become available to plants in the form of NH_3. It also produces phytohormones (i.e., IAA, cytokinins, and other compounds). Plants will provide carbon or sugars and amino acids, namely sugars, mainly sucrose and glucose, for endophytic bacteria (Gaiero et al., 2013).

Bacteria Endophytes on NPK Nutrient Concentration

N and P leaf significantly increased with APE35 application compared to the control but was not different from other endophytes. In contrast, the treatment of endophytic bacteria did not affect the nutrient K concentration (Table 4). Endophytic bacterial isolates produced auxin, ammonia, and dissolved phosphate (Fig. 1). Four isolates of endophytic bacteria tested were able to produce N in the urease test. APE35 was able to solubilize phosphate. Gusmaini et al. (2019) found that in bitter plants, endophytic bacteria could provide nutrients for the growth of host plants.

Table 3. Effect endophyte to root fresh weight, head diameter, and dry head weight

Endophytic bacteria isolate	Parameter		
	Root fresh weight (g)	**Head diameter (cm)**	**Dry head weight (g)**
Control	5.86^{b}	45.04^{b}	31.97^{c}
APE35	6.37^{ab}	51.21^{ab}	40.37^{a}
PTM3	6.73^{a}	54.33^{a}	39.57^{ab}
APE35+BAT	5.98^{b}	46.63^{b}	34.79^{abc}
APE35+BAT+EQ26	5.92^{b}	48.50^{ab}	32.65^{bc}

Remarks: The numbers followed by the same letter are not significantly different from the 5% HSD test

Table 4. Effect endophyte to NPK nutrient concentration

Endophytes	NPK leaf concentration (%)		
	N total	**P**	**K**
Control	3.268^{b}	0.640^{b}	7.233^{a}
APE35	3.716^{a}	0.802^{a}	7.171^{a}
PTM3	3.506^{ab}	0.628^{b}	6.567^{a}
APE35 and BAT	3.540^{ab}	0.714^{ab}	6.837^{a}
APE35, BAT, and EQ26	3.538^{ab}	0.677^{ab}	6.878^{a}

Remarks: The numbers followed by the same letter in the same column are not significantly different from the 5% HSD test

Celery Varieties or Endophytic Bacteria on Total Flavonoids

The tested secondary metabolites in celery were total flavonoid concentrations. Amigo and Aroma have significantly higher total flavonoid concentrations than Summer Green. While the endophytic bacteria APE35 and PTM3 significantly affected the total flavonoid concentration, which was higher than the control. Based on control, the content of flavonoid due to PTM3 and APE35 endophytes increased by 15.18 and 14.17%, respectively. APE35 and BAT, the combination of APE35, BAT, and EQ26 endophytes, have content flavonoid concentrations of 9.98 and 11.67%, respectively, compared to the control, but did not differ from all the endophytic bacterial isolates treatments (Table 5). Production of secondary metabolites can be increased due to the role of endophytic bacteria. The existence of endophytic bacteria stresses celery plants to increase the total flavonoid concentration. Secondary metabolites act as stress, biotic and abiotic (Angin et al., 2019). Endophytic bacteria can produce secondary metabolites like secondary metabolites produced by their host. APE35 and PTM3 significantly increased total flavonoids than the control but were not different from the endophytic combination of APE35 and BAT and the combination of APE35, BAT, and EQ26. Based on Herlina et al. (2016), IAA-producing bacteria play a role in cell enlargement, stimulate abscission, and form tissue in xylem and phloem to affect plant development. Nurchayanti et al. (2019) stated that IAA would be converted into an amino acid, namely tryptophan, which will then be converted back by bacteria into other amino acid products and used for metabolism.

The total concentration of flavonoids is a secondary metabolite that includes antioxidants derived from natural phenolic compounds. Based on the study results, Amigo and Aroma varieties were upland varieties with a significantly higher total flavonoid concentration than the Summer Green which is a lowland variety. Each variety of celery has a different phytochemical ability to respond to a growing environment that is not suitable such as high temperatures and low humidity. Plants can be more tolerant to high temperatures, thereby increasing the content of secondary metabolites as adaptation mechanism (Du et al., 2013). In addition to plants, varieties release chemical compounds to adapt to less-than-optimal growing environmental conditions. Microorganisms such as endophytic bacteria also affect the prodution of chemical compounds, namely total flavonoids and antioxidants, to protect other from organisms and unfavorable ecological conditions.

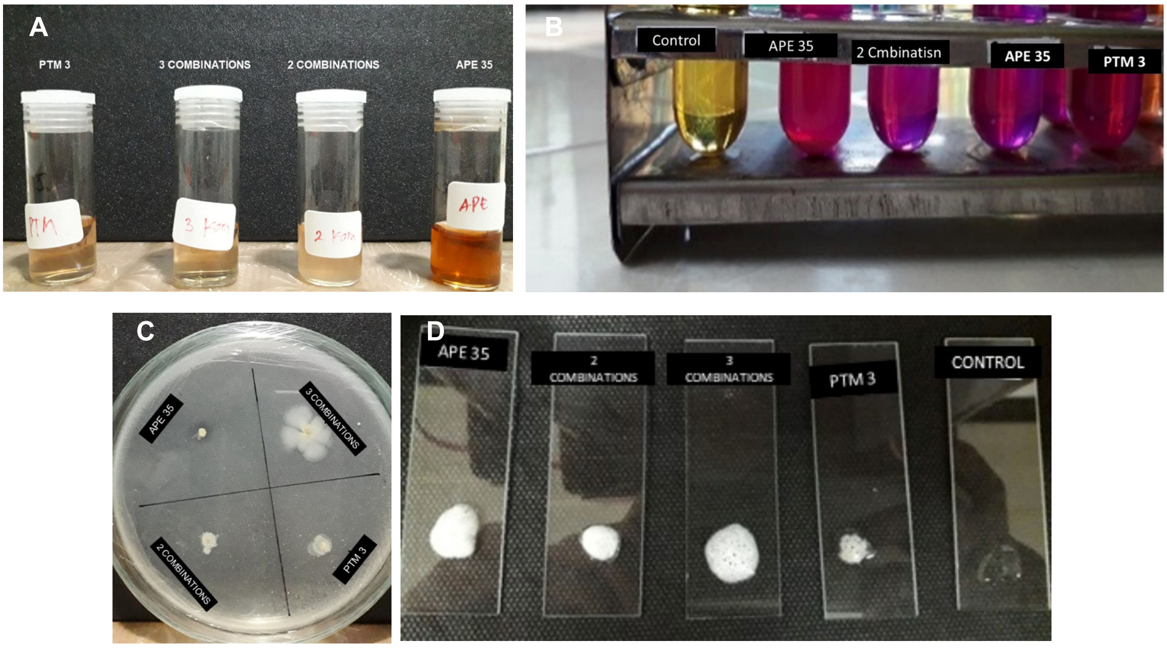

Fig. 1. Qualitative physiological test using IAA (A). urea base agar (B). phosphate solvent (C). and catalase (D). text in the object could not see clearly

Table 5. Total flavonoid content of celery varieties under different endhopytic bacteria treatments

Varieties	Total flavonoids (mg QE/100g)
Amigo	1506.783[a]
Aroma	1600.302[a]
S. Green	1327.976[b]
Endophytes	**Total flavonoids (mg QE/100g)**
Control	1274.354[b]
APE35	1484.818[a]
PTM3	1502.459[a]
APE35 and BAT	1415.679[ab]
APE35, BAT, and EQ26	1442.789[ab]

Remarks: The numbers followed by the same letter are not significantly different from the 5% HSD test

Table 6. Effect of endophytes on anthocyanin compounds

Endophyte	Anthocyanin (mg/cm^2)
Control	0.0326[b]
APE35	0.0419[a]
PTM3	0.0390[ab]
APE35 and BAT	0.0386[ab]
APE35, BAT, and EQ26	0.0373[ab]

Remarks: The numbers followed by the same letter are not significantly different from the 5% HSD test

Table 7. Carotenoids concentration of celery varities under different endophytoc bacteria treatments

Varieties	Carotene (mg/cm^2)
Amigo	0.01085[a]
Aroma	0.01026[ab]
S. Green	0.00960[b]
Endophyte	**Carotene (mg/cm^2)**
Control	0.01002[ab]
APE35	0.01117[a]
PTM3	0.01082[a]
APE35 and BAT	0.00940[b]
APE35, BAT, and EQ26	0.00977[ab]

Remarks: The numbers followed by the same letter are not significantly different from the 5% HSD test

Endophytic Bacteria on Celery Anthocyanins and Carotenoids

APE35 has a significantly higher anthocyanin concentration than the control but was not different from other endophytic bacteria treatments (Table 6). Carotenoids are organic pigments found in plants and other organisms like algae and bacteria. The vital role of carotene in physiological processes in plants is to produce antioxidants and preserve the ongoing process of photosynthesis. Based on Table 7, the effect of carotene on the celery Amigo was significantly higher than the Summer Green and not different from the Aroma. APE35 endophytic bacteria isolates and PTM3 consortium isolates on the carotene content were significantly higher and not different from the control and the combination of three isolate treatments. Endophytic bacterial isolates from the two combinations of APE35 and BAT were significantly lower than the control in increasing the content of carotenoid compounds. The combination of the two isolates may be incompatible or commensal, or parasitic. One isolate of endophytic bacteria did not increase and had a negative impact on carotene compounds.

Anthocyanins and carotenoids are pigments and strong antioxidants (Clemente et al., 2021) in a plant that assisted chlorophyll in photosynthesis (Waldemar et al., 2020). Anthocyanins are included in the type of flavonoid because they contain antioxidants and are included in the polyphenol group. The utilization of anthocyanins in various organisms can encourage various physiological functions in plants. Anthocyanins in plants are used as coloring agents, protecting plants from biotic and abiotic stresses, and as photoprotection against UV-B radiation. In humans, anthocyanins are used as bioactive compounds to prevent various chronic diseases. Anthocyanins are used as additives in food and beverage ingredients in the food sector. While in the industrial sector, anthocyanins are used in the manufacture of cosmetics. Anthocyanins also function as attractants or insect traps in flowers that provide attractive colors to help pollinate due to insect activity. Based on the data obtained, the effect of endophytic bacteria on the increase in anthocyanins occurred in single isolate APE35, while the combination isolates and PTM3 showed the same results. Endophytic bacteria can increase compounds or products produced by the plant (Okungbowa et al., 2019). The same results also occur in carotenoid chemical compounds in celery plants. Amigo and Arona showed higher compounds than the lowland variety, Summer Green. Amigo and Aroma are highland varieties that experience abiotic stress in a less optimal environment when grown in lowland. They produce secondary metabolites to adapt and tolerate environmental conditions in the lowlands. This result also affects other secondary metabolite compounds like flavonoid compounds.

Interaction of Varieties and Endophytic Bacteria in Celery Plant

Summer green with endophytic bacteria or the Amigo with PTM3 has a significantly higher leaf area than other interactions (Table 8). The number of tillers at 8 WAP of PTM3 endophytic bacteria significantly increased Amigo and Summer Green compared to other interactions (Table 9). On 8 WAP, the interaction of Amigo and Aroma with PTM3 and Aroma with endophytic bacteria with three combinations of APE35, BAT, and EQ26 on number of leaflets were significantly greater than other interactions (Table 10). On total chlorophyll, the interaction between Amigo and PTM3 varieties was significantly higher than the control and not different from the interaction between the lowland variety Summer Green and the endophytic bacteria PTM3 and with other treatment interactions (Table 11). Total chlorophyll captures sunlight and converts water and carbon dioxide, which produces oxygen, and the product is glucose ($C_6H_{12}O_6$). This process is proven to increase plant productivity, as indicated by the total fresh weight (Malik et al., 2021). APE35 or PTM3 with Amigo produced significantly higher head fresh weight (78.529 and 75.054 g/plant), and total fresh weight (81.67 and 85.395 g/plant) than Summer Green when without endophytic bacteria (Table 12 and Table 13). The host-endophyte relationship can be regarded as a flexible, dynamic interaction, in which endophytic bacteria alter their gene expression or produce different metabolites based on small changes in host plant growth and vice versa (Ek-Ramos et al., 2019). This phenomenon could be seen in different responses of different celery varieties to the different endophytic bacteria.

APE35 single isolate was a pure isolate from one test bacteria isolate, while the combined isolate came from several endophytic bacteria. PTM3 bacterial isolates were consortium isolates from a mixture of bacterial populations in communities that naturally possibly had cooperative, commensal, and mutualistic relationships. They have a synergistic relationship between bacteria and do not inhibit the growth of each other (Asri & Zulaika, 2016). Endophytic bacterial physiology proved this for all IAA isolates. Based on the research of Herlina et al. (2016), IAA-producing bacteria play a role in cell enlargement, stimulate abscission, and form tissues in the xylem and phloem that affect plant development. IAA phytohormones will be converted into amino acids, namely, tryptophan, which bacteria will then be converted back into other amino acid products and used for metabolism (Nurchayanti et al., 2019).

Table 8. Interaction varieties and bacteria endophyte on leaf area (cm^2)

Varieties	Endophytes				
	Control	APE35	PTM3	APE35 and BAT	APE35, BAT, and EQ26
Amigo	447.00^{d}	527.13^{bcd}	72.73^{ab}	475.097^{cd}	469.95^{cd}
Aroma	423.27^{d}	622.60^{abcd}	557.86^{abcd}	480.18^{cd}	557.31^{abcd}
S. Green	466.13^{cd}	784.44^{a}	765.06^{a}	755.56^{ab}	682.79^{abc}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test

Table 9. Interaction of varieties and bacteria endophyte on tiller number

Days to	Varieties	Control	APE35	PTM3	APE35 and BAT	APE35, BAT, and EQ26
4 WAP	Amigo	1.0^{c}	1.0^{c}	2.50^{a}	1.0^{c}	1.8^{abc}
	Aroma	1.0^{c}	1.0^{c}	2.0^{ab}	1.3^{bc}	2.0^{ab}
	S.Green	1.0^{c}	1.5^{bc}	2.0^{ab}	1.8^{abc}	1.8^{abc}
6 WAP	Amigo	1.2^{d}	1.8^{bcd}	2.6^{a}	1.6^{bcd}	2.1^{ab}
	Aroma	1.2^{d}	1.9^{bcd}	1.9^{bcd}	1.8^{bcd}	2.0^{abc}
	S.Green	1.3^{cd}	1.4^{bcd}	1.9^{abc}	1.8^{bcd}	1,9^{bc}
8 WAP	Amigo	1.4^{c}	1.9^{bc}	3.3^{a}	1.7^{bc}	2.3^{b}
	Aroma	1.4^{c}	2.1^{bc}	2.0^{bc}	2.0^{bc}	1.9^{bc}
	S.Green	1.4^{c}	2.0^{bc}	2.5^{ab}	1.9^{bc}	1.9^{bc}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test; WAP = week after planting

Some variables observed in this research contributed as the yield components to the total fresh weight and flavonoids. The yield components are plant height, number of stalks, number of leaflets, leaf area, chlorophyll, anthocyanins, and carotenoids (pigments), and crown diameter. Extensive roots shown by the root weight will make the plant make plants able to take soil nutrients better. The plant's N, P, and K act as minerals needed by the plant to produce plant biomass in the plant metabolism. The high contribution of each of these variables will increase crop yields. The higher the photosynthate induced higher the flavonoids because it encourages further metabolism.

Table 10. Interaction varieties and bacteria endophyte on total leaflet number

Varieties	Endophytes				
	Control	**APE35**	**PTM3**	**APE35 and BAT**	**APE35, BAT, and EQ26**
Amigo	37.40^{i}	45.75^{fgh}	62.70^{a}	43.87^{hi}	53.34^{cde}
Aroma	48.72^{efgh}	53.92^{cde}	61.47^{ab}	48.67^{efgh}	59.95^{abc}
S.Green	44.87^{gh}	55.75^{bcd}	51.75^{def}	50.75^{defgh}	49.00^{efgh}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test

Table 11. Interaction of varieties and endophytic bacteria on total chlorophyll (mg/cm^{2})

Varieties	Endophytes				
	Control	**APE35**	**PTM3**	**APE35 and BAT**	**APE35, BAT, and EQ26**
Amigo	0.0398^{b}	0.0525^{ab}	0.0600^{a}	0.0457^{ab}	0.0461^{ab}
Aroma	0.0405^{b}	0.0479^{ab}	0.0447^{b}	0.0436^{b}	0.0479^{ab}
S.Green	0.0393^{b}	0.0436^{b}	0.0474^{ab}	0.0438^{b}	0.0429^{b}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test

Table 12. Interaction of varieties and endophytic bacteria on fresh head weight (g/plant)

Varieties	Endophytes				
	Control	**APE35**	**PTM3**	**APE35 and BAT**	**APE35, BAT, and EQ26**
Amigo	50.870^{abc}	78.529^{a}	75.054^{ab}	57.214^{abc}	46.324^{c}
Aroma	52.823^{abc}	60.095^{abc}	67.921^{abc}	51.378^{abc}	49.815^{bc}
S.Green	52.415^{abc}	59.511^{abc}	63.305^{abc}	64.308^{abc}	67.224^{abc}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test

Table 13. Interaction of varieties and endophytic bacteria on total plant fresh weight (g/plant)

Varieties	Endophytes				
	Control	**APE35**	**PTM3**	**APE35 and BAT**	**APE35, BAT, and EQ26**
Amigo	51.960^{c}	81.671^{ab}	85.395^{a}	62.755^{abc}	56.458^{abc}
Aroma	55.519^{bc}	66.108^{abc}	74.189^{abc}	58.822^{abc}	58.750^{abc}
S.Green	58.471^{abc}	65.995^{abc}	69.354^{abc}	70.957^{abc}	73.404^{abc}

Remarks: The numbers followed by the same letter in the same row and column are not significantly different from the 5% HSD test

CONCLUSION

Highland celery varieties can be grown in the lowlands with the supplemental application of endophytic bacteria to increase their production. Amigo is short but has a significantly higher stalk number, carotenoids, and flavonoids than Summer Green (lowland control). Endophytic bacteria increased plant growth, N, P, pigments, and flavonoids. Amigo applied with APE35 or PTM3 produced the best yield (81.67 and 85.395 g/plant) and total flavonoids (1484.818 and 1502.459 mg QE/100g) for highland celery when planted in.

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