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Research article

The Effect of Seed Soaking with Suspensions of Pseudomonas Alcaligenes and Bacillus on the Growth and Yield of Bitter Melon (Momordica Charantia L.) in a Greenhouse

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Abstract.
The aim of this research was to determine the impact of soaking bitter melon seeds in a *Pseudomonas alcaligenes* and *Bacillus* sp bacteria suspension on the growth and yield of bitter melon. A randomized block design was used with 11 treatments consisting of three isolates of *P. alcaligenes*, four isolates of *Bacillus* sp, one mixed suspension of *P. alcaligenes*, one mixed suspension of *Bacillus* sp, one mixed suspension of *P. alcaligenes* with *Bacillus* sp, and one control, each of which was repeated four times for a total of 44 experimental pots. Data were analyzed using a variance test with one-way ANOVA. The results showed that the highest number of fruits was obtained from the *Bacillus* sp2 treatment (4.25 units), followed by the mixed *Bacillus* sp, *Bacillus* sp3, and *P. alcaligenes* TmA1 (4.00 units), while the control only yielded 3.25 units. The highest fresh weight per fruit was obtained from the *P. alcaligenes* TrN2 treatment (89.25 g), followed by *Bacillus* sp2 (87.2 g), and mixed *Bacillus* sp (80.66 g), which was an increase of 34.57%, 31.48%, and 21.62% over the control, respectively.

Keywords: seed soaking; bitter melon; suspension of *Bacillus sp.*; *P. alcaligenes*

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1. Introduction

Bitter gourd is a fruit vegetable plant from the Cucurbitaceae family, classified as a herbaceous plant aged one year or more, growing, creeping, and climbing. Bitter melon was not in demand in the past, so it was not appropriately cultivated due to low consumer demand. However, bitter melon is increasingly popular because its vitamin content is increasingly recognized, and there are new varieties with fruit flavors and appearances that suit consumer tastes. Bitter gourd fruit is a good source of vitamin C, vitamin A, phosphorus, and iron. In addition, the bitter gourd stem tip is a good source of pro-vitamin A, protein, thiamine, and vitamin C [1]. The increasing demand for bitter melon in

agribusiness in Indonesia impacts the need for bitter melon seeds, which has increased very significantly.

Although the prospect of the bitter melon market is quite bright, the cultivation of bitter melon at the farmer level is still a side business, on a small scale, and without intensive maintenance, so that growth is still not optimal. On the other hand, organic crop cultivation is a commodity that has promising prospects. Therefore, efforts are needed to increase yields through various actions, including proper, effective, and efficient cultivation techniques. In addition, as far as possible in cultivating bitter melon plants can reduce the chemical fertilizers use and synthetic chemical pesticides.

One of the efforts made to maintain and increase plant production is PGPR (Plant Growth Promoting Rhizobacteria). PGPR is soil microbes in rhizosphere that can either directly or indirectly, affected plants growth and development [2]. PGPR function is to restore soil fertility because some bacteria from the PGPR group are nitrogen-fixing bacteria such as the genus Rhizobium and Azotobacter, and phosphate solubilizing bacteria such as the genus Arthrobacter, Bacterium, and Mycobacterium [3]. The results of Widnyana and Javandira research claimed that the treatment of *Bacillus* sp. to tomato seeds for 10-30 minutes can stimulate tomato seed germination. [4]. Similar investigations that immerse seed treatment with *P. alcaligenes* showed 25% faster germination for swamp cabbage encouraged, increased in performance by 24.4% [5], while Taufik stated the use of PGPR on chili seeds was able to enhanced the vegetative and generative growth of chili plants [6]. The positive effect of *Pseudomonas spp.* can occupy plant root tissue surface and supply essential nutrients. In addition, several bacteria can interrupt the root as endophytes without changes in plants morphology [7].

Maunuksela in 2004 stated that the rhizobacteria of the *Pseudomonas fluorescens*, *Bacillus sp.*, and *Serratia spp.* groups could generate growth hormones like indole acetic acid (IAA), which can enhanced plant growth [8]. Low seed germination can inhibit plant growth, causing abnormal root growth so that the roots cannot absorb water and nutrients optimally. One of the efforts that can be done is by providing PGPR as a natural growth promoter. The results of previous studies stated that tomato seed immerse treatment with a suspension of *P. alcaligenes* and *Bacillus sp*. was able to increase 10.54% of leaves number, 4.04% of stem fresh weight, 55.16% of plant height, 162.83% of fruit number, 344.44% of fruit weight, and 17.90% of root length than the control [9].

This study used PGPR bacteria, namely *P. alcaligenes* and *Bacillus sp*., with the hope that it can be an alternative in stimulating germination, growth, and increasing the yield

of bitter melon. The aim of this research is to analyze the impact of *Bacillus sp.* and *Pseudomonas alcaligenes* when given alone or after being combined on the yield and growth of bitter melon plants.

2. Methodology

2.1. Tools and materials

The materials used in this research included: *Pseudomonas alcaligenes* (TmA1, KtS1, TrN2), *Bacillus sp* (1, 2, 3, 4), seeds of bitter melon varieties LIPA F1, and growing media. The tools used include Laminar airflow, 90% alcohol, autoclave, oven, scales, Bunsen lamp, stove, ruler, enkase, polybag, bucket, hoe, Petri dish, tweezers, scissors, plastic, needle ose, measuring cup, bamboo, strap, camera, translucent clear plastic, and stationery.

2.2. Research Design and Treatment

Randomized Block Design (RBD) was used with 11 treatments which were repeated four times. The treatments carried out in this study were: P0 (control), (P1) = *P. alcaligenes* TmA1, (P2) = *P. alcaligenes* TrN2, (P3) = *P. alcaligenes* KtS1, (P4) = mixed *P. alcaligenes* (P5) = *Bacillus sp.*1., (P6) = *Bacillus sp 2*, (P7) = *Bacillus sp.*3, (P8) = *Bacillus sp.* 4, (P9) = mixed *Bacillus sp*., and (P10)= mixed *P. alcaligenes* with *Bacillus sp*.

2.3. Preparation of Bacterial Isolates of P. alcaligenes and Bacillus sp

P. alcaligenes and *Bacillus sp*. are from the Laboratory of Agrotechnology, Faculty of Agriculture, Mahasaraswati University Denpasar, have previously been studied and identified, especially for *P. alcaligenes*. The bacterial suspensions of *P. alcaligenes* and *Bacillus sp.* were isolated on NB (Nutrien Broth) media in 100 ml Erlenmeyer and cultured for 48 hours to be ready for application.

2.4. Bacterial suspension treatment and planting bitter melon seeds

Bitter gourd seeds were soaked for 20 minutes with suspension isolate according to the treatment that had been determined using a Petri dish as a soaking container. After

the soaking, the seeds are drained and stored in a protected container in a shady place and ready to be sown/planted on the planting media that has been prepared in polybags. Planting media in the form of soil and compost media (3:1) that have been opened to minimize contaminants. Each polybag is filled with 5 kg of planting media. After the bitter gourd seeds were 1 and 2 weeks old, they were watered with 100 ml of rhizobacteria isolate suspension.

2.5. Variable Observation and data analysis

Observations were made on the following variables: germination time, number of leaves, plant height, fruits number, fruit fresh weight, roots fresh weight, fresh weight of stems, fresh plant weight, dry fruit weight, root dry weight, dry stem weight, weight plant dryness, fruit length, root length. Data was analyzed using the ANOVA test and for the average difference, test using the 5% and 10% LSD tests [10].

3. Results and Discussion

Statistical analysis result on the observed parameters, showed there a strong significant effect ($P<0.01$) by treatment on the parameters of fruit number, length of fruit, weight of fresh fruit, fruit oven-dry weight, length of root, and fresh weight, and oven-dry weight stems, but had no significantly effect ($P>0.05$) on the overy weight of bitter melon roots.

3.1. Bitter melon seed germination duration

Figure 1 shows the germination rate of bitter melon seeds in each seed soaking treatment. The fastest germination of bitter melon seeds occurred in immersion with bacterial suspension of *P. alcaligenes* TmA1, mixed *P. alcaligenes*, and *Bacillus sp*.4 with an average of 3.5 days. Germination within four days occurred when bitter melon seeds were soaked in suspension with *P. alcaligenes* TrN2, *Bacillus sp*.1, *Bacillus sp*.2, and *Bacillus sp*.3. Bitter gourd seed germination within five days was found by immersing seeds with *P. alcaligenes* KtS1, mixed *Bacillus sp.*, mixed *P. alcaligenes* with *Bacillus sp*., and controls.

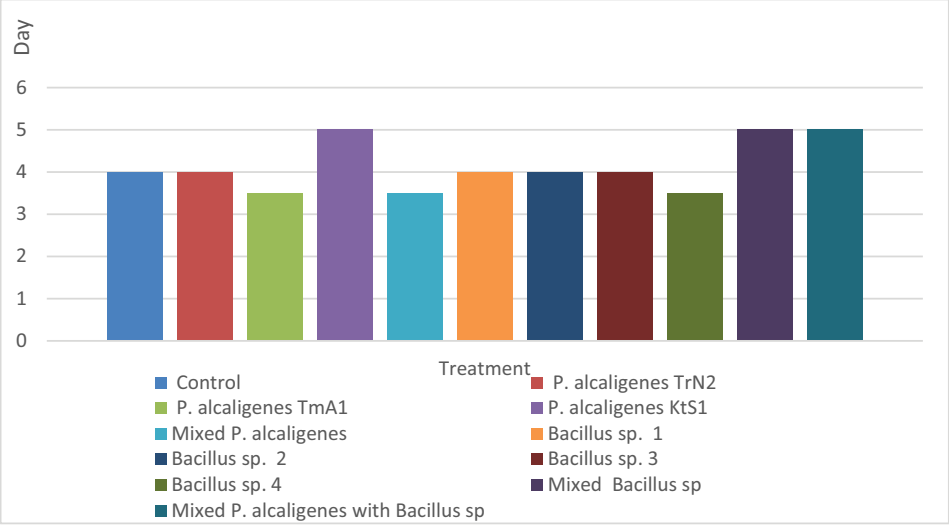

Figure 1: Effect of soaking period bitter melon seeds on germination speed.

3.2. Fruit variable

Table 1 shows the average number of bitter melon fruit, fruit length, fruit fresh weight, and oven-dry weight of bitter melon. The highest number of fruits was found in *Bacillus sp.* 2, namely 4.25 pieces, but not significantly different than *Bacillus sp.* 3, mixed *Bacillus sp.*, and *P. alcaligenes* TmA1 treatment. The minimum number of fruits, namely 3 pcs, was found in the treatment of *P. alcaligenes* KtS1, not significantly different from the control, *P. alcaligenes* TmA1, mixed *P. alcaligenes*, *Bacillus sp.*, and mixed *Bacillus sp.* The most extended bitter melon fruit was found in the treatment of *P. alcaligenes* TrN2, which was 24.35 cm, not significantly different from the mixed *Bacillus sp.* (22.68 cm) and *Bacillus sp.* 2 (22.51 cm). The shortest bitter melon found in the control treatment (17.53 cm) was not significantly different from the treatment of *Bacillus sp.*4 (17.91 cm), *P. alcaligenes* KtS1 (17.66), *P. alcaligenes* TmA1 (18.11 cm), *Bacillus sp.4* (18.55 cm), and *Bacillus sp.3* (18.75 cm).

Notes :

1. The same letter behind the average value shows a non-significant difference (P>0.05)
2. The different letters behind the average value shows a significant difference (P<0.05) to very significant (P<0.01)

The highest fresh weight of bitter melon was showed on the treatment of *P. alcaligenes* TrN2, which was 89.25 g, not significantly different from *Bacillus sp.*2 (87.20 g), and Mixed *Bacillus sp.* (80.66 g), but significantly different from all other treatments. The

TABLE 1: The average number of fruit, fruit length, fresh weight, and oven-dry bitter melon on PGPR bacterial treatment.

No	Treatments	Number of fruit (unit)	fruit length (cm)	fresh weight (g)	oven-dry weight (g)
1	Control	3.25 bc	17.53 c	66.32 d	6.02 f
2	*P. alcaligenes* TmA1	3.25 bc	18.11 c	65.75 d	6.30 ef
3	*P. alcaligenes* TrN2	4.00 a	24.35 a	89.25 a	9.01 a
4	*P. alcaligenes* KtS1	3.00 c	17.66 c	79.98. b	7.37 d
5	Mixed *P. alcaligenes*	3.25 bc	20.99 b	81.00 b	7.99 c
6	*Bacillus sp.* 1	3.25 bc	18.55 c	67.49 d	6.18 ef
7	*Bacillus sp.* 2	4.25 a	22.51 ab	87.20 a	8.56 b
8	*Bacillus sp.* 3	4.00 a	18.75 c	73.78 b	7.05 d
9	*Bacillus sp.* 4	3.25 bc	17.91 c	62.55 d	6.54 e
10	Mixed *Bacillus sp*	4.00 a	22.68 ab	80.66 a	8.42 b
11	Mixed *P. alcaligenes* with *Bacillus sp*	3.50 b	18.36 c	73.57 c	7.13 d
	LSD 5%	0.56	2.05	5.45	0.40
	LSD 1%	0.75	2.76	7.33	0.54

lowest fresh fruit weight was found in control (66.32 g), not significantly different from the treatment of *P. alcaligenes* TmA1 (65.75 g), *Bacillus sp.* 1 (67.49 g), and *Bacillus sp.*4 (62.55 g). *P. alcaligenes* TrN2 treatment (9.01 g) showed the highest oven-dry weight of bitter melon and showed significantly different than others, while the lowest oven-dry weight was found in control (6.02 g), not significantly different from the *P. alcaligenes* TmA1 treatment. (6.30 g), and *Bacillus sp.* 1 (6.18 g), but significantly different from all other treatments.

3.3. Root variable

Statistical analysis result showed that root dry weight had no significant effect (P>0.05) by treatment application but significantly (P<0.05) to very significant (P<0.01) on root length and fresh root weight. The longest bitter melon roots were found in the Mixed *P. alcaligenes* treatment (89.59 cm), not significantly different from the *P. alcaligenes* TmA1 (84.63 cm), *P. alcaligenes* TrN2 (89.62 cm), and Mixed *Bacillus sp*. (81.14 cm) treatments. The shortest bitter melon roots were found in *Bacillus sp.* 3 (63.25 cm), and not significantly different from the *Bacillus sp.*1 (66.41 cm), *Bacillus sp.*4 (68.75 cm), Mixed *P. alcaligenes* with *Bacillus sp*. (67.25 cm), *P. alcaligenes* KtS1 (63.50 cm), and control (66.68 cm). The best root fresh weight was showed in *P. alcaligenes* TrN2 (11.92 g) treatment and was significantly different than others. The lowest fresh root weight was found in control (7.16 g), not significantly different from *P. alcaligenes* KtS1 treatment (7.56

g), *Bacillus sp.* 1 (6.80 g), *Bacillus sp.*2 (8.43 g), and *Bacillus sp*.4 (6.98 g). Specifically on the oven-dry weight of bitter melon root, PGPR treatment had no significant impact.

Table 2: Average root length of bitter melon, fresh root weight, and oven-dry weight of bitter melon root on PGPR bacterial treatment.

No	Treatments	Root length (cm)	Fresh root weight (g)		Root oven-dry weight (g)
1	Control	66.68 c	7.16	ef	1.16 a
2	*P. alcaligenes* TmA1	84.63 ab	9.76	b	1.01 a
3	*P. alcaligenes* TrN2	89.62 ab	11.92	a	1.53 a
4	*P. alcaligenes* KtS1	63.50 c	7.56	cdef	1.03 a
5	Mixed *P. alcaligenes*	89.59 a	8.82	bcd	1.52 a
6	*Bacillus sp.* 1	66.41 c	6.80	f	1.74 a
7	*Bacillus sp.* 2	79.00 b	8.43	bcde	1.89 a
8	*Bacillus sp.* 3	63.25 c	8.67	bcd	1.24 a
9	*Bacillus sp.* 4	68.75 c	6.98	ef	1.18 a
10	Mixed *Bacillus sp*	81.14 ab	8.97	bc	2.02 a
11	Mixed *P. alcaligenes* and *Bacillus sp*	67.25 c	7.37	def	1.12 a
	LSD 5%	19.64	1.46		1,29
	LSD 1%	26.44	1.97		1,73

Notes :

1. The same letter behind the average value shows a non-significant difference (P>0.05)
2. The different letters behind the average value shows a significant difference (P<0.05) to very significant (P<0.01)

3.4. Bitter melon plant stem variable

Statistical analysis result showed that the PGPR treatment had strong significant effect (P<0.01) on the fresh and oven-dry weight of bitter melon stems. The highest stem fresh weight was showed in *P. alcaligenes* KtS1 treatment (74.35 g), not showed significant different than *P. alcaligenes* TrN2 treatment (72.50 g), Bacillus sp. 4 (71.57 g), *Bacillus sp.*1 (67.15 g), Mixed *P. alcaligenes* (63.55 g), and *Bacillus sp*.2 (62.82 g). The lowest fresh stem weight of bitter melon was found in the treatment of *P. alcaligenes* KtS1 (74.35 g), not significantly different from *Bacillus sp.3* (52.14 g), Mixed *Bacillus sp.* (55.00 g), control (59.00 g), Mixed *P. alcaligenes* with *Bacillus sp.* (60.00 g), Mixed *P. alcaligenes* (63.55 g), and *Bacillus sp.*2 (62.82 g). The treatment of *P. alcaligenes* TrN2 (7.99 g) was performed highest stem oven-dry weight meanwhile not significantly different than the

treatments of *P. alcaligenes* KtS1 (7.16 g), *Bacillus sp.*4 (7.42 g), Mixed *Bacillus sp*. (6.52 g), and Mixed *P. alcaligenes* with *Bacillus sp*. (6.60 g). The lowest oven-dry weight of bitter melon stems was found in Mixed *P. alcaligenes* (5.15 g), not significantly different from *Bacillus sp.*3 (5.56 g), control (5.81 g), *P. alcaligenes* TmA1 (54.00 g), *Bacillus sp.*2 (6.15 g), Mixed *Bacillus sp.* (6.52 g), and Mixed *P. alcaligenes* with *Bacillus sp*. (6.60 g)

TABLE 3: Average fresh weight of stems and oven-dry weight of bitter melon in each treatment with PGPR bacteria.

No	Treatments		fresh weight (g)		oven-dry weight (gr)	
1	Control		59.00	cde	5.81	bcd
2	*P. alcaligenes* TmA1		54.00	e	6.06	bcd
3	*P. alcaligenes* TrN2		72.50	ab	7.99	a
4	*P. alcaligenes* KtS1		74.35	a	7.16	abc
5	Mixed *P. alcaligenes*		63.55	abcde	5.15	d
6	*Bacillus sp.* 1		67.15	abcd	7.11	abc
7	*Bacillus sp.* 2		62.82	abcde	6.15	bcd
8	*Bacillus sp.* 3		52.14	e	5.56	cd
9	*Bacillus sp.* 4		71.57	abc	7.42	ab
10	Mixed *Bacillus sp*		55.00	de	6.52	abcd
11	Mixed *P. alcaligenes* with *Bacillus sp*		60.00	bcde	6.60	abcd
	LSD 5%		12.58		1.83	
	LSD 1%		16.94		2.47	

Notes :

1. The same letter behind the average value shows a non-significant difference (P>0.05)
2. The different letters behind the average value shows a significant difference (P<0.05) to very significant (P<0.01)

PGPR is a consortium of actively colonize bacteria in rhizosphere and contribute to increasing plant yield and soil quality [11]. Principle of giving PGPR is to improve amount of active bacteria around plant roots to provide beneficial effect for plants. The advantages of using PGPR are increasing mineral content and nitrogen fixation, increasing plant tolerance to environmental stress as a biofertilizer, protecting plants from plant pathogens, and stimulate indole-3-acetic acid (IAA) production [12].

According to Tenuta, PGPR mechanism in provide plant health occurs in three ways, namely: a). Suppress the development of pests/diseases (bioprotectants), have a direct effect on plants in dealing with diseases and pests, b) produce phytohormones (biostimulants), IAA; cytokinins; gibberellins; and inhibit ethylene production, can increasing fine

roots surface area, c) increase nutrients availability for plants (biological fertilizers) [13]. However, according to Mc. Milan, PGPR is important in promoting plant growth include: (a) increasing fixation of nitrogen in legumes, (b) increasing the nitrogen-fixing bacteria population (c) increasing sulfur, phosphorus, copper, and iron (d) hormone production, (e) increasing beneficial fungi or bacteria population, (f) suppress pathogenic fungi, pathogenic bacteria, and insect pests [14][15].

The ability to suppress the growth of soilborne pathogens by PGPR depends on its ability to produce siderophores. Widnyana's research results showed that there were 3 isolates of *P. alcaligenes*, namely *P. alcaligenes* KtS1, *P. alcaligenes* TrN2, and *P. alcaligenes* TmA1 which were antagonistic to *Fusarium oxysporum* f. sp. *lycopersici* with 71.17% inhibition on *P. alcaligenes* KtS1, 80.53% on *P. alcaligenes* TrN2, and 79.22% on *P. alcaligenes* TmA1. Three bacterial were also able to form siderophores of 0.239 (*P. alcaligenes* KtS1), 1,320 (*P. alcaligenes* TrN2), 1,467 (*P. alcaligenes* TmA1) absorbance at 500 nm [16].

Rahni suggested that PGPR can produce cytokinins, IAA, gibberellins, abscisic acid, and ethylene, where IAA is the active form of the auxin hormone found in plants and can develop crop and yields quality. The functions of the IAA hormone for plants include increasing cell development, stimulating new roots formation, promoting plant growth, encouraging flowering, and increasing enzyme activity [17]. Egamberdiyeva also reported that IAA and nitrogenase enzymes were shown to improve nutrient uptake of maize plants and dry weight [18]. Microbes can produce IAA that enhance growth and elongation of root so that more nutrient can absorbed from the soil through expansive root surface [19].

Giving PGPR to plants can also increase plant resistance to pathogens. The increase in plant resistance was indicated by an increase in the total salicylic acid and phenol levels in plant tissue after being given induction treatment with PGPR. Pseudomonas sp. is one of the bacteria that is widely studied and used as a PGPR and biocontrol agent of the disease [20][21]. Pseudomonas spp. was reported as a biological control agent of [22], vanilla stem rot disease control [23] and tomato wilt disease [15].

The results of other studies showed that the *Pseudomonas spp.*W80 treatment could increase the total phenol level in the treatment, which averaged 260.27 mg/g or improve of 106.89% than control (distiled water). In *Pseudomonas spp.* W02 treatment, there was an increase in the level of salicylic acid by 37.65 ppm or a 72.94% increasing than the control. *Pseudomonas spp*. bacteria W02, W68, and W80 have different abilities to protect plant of vanilla from pathogens. Adding *Pseudomonas spp*. suspension to

plants of vanilla such as stems, roots, or leaves can improve vanilla plants systemic resistance by performance of salicylic acid and phenol in plant of vanilla tissues [23].

Salicylic acid is a produced naturally phenolic compound in plants and is a significant component of local and systemic plant resistance. Colonization of tobacco root with P. fluorescens strain CHAO and Tobacco Mozaic Virus TMV in the leaves caused salicylic acid enhancement in the leaves [24]. Salicylic acid can induce systemic resistance after transported to the leaves. *Pseudomonas spp*.W02, W68, and W80 can protect vanilla plant from pathogens [25]. Fluorescent *Pseudomonas spp*. effective suppressed soilborne plant diseases through play siderophore-mediated competition for iron, lytic enzymes production, antibiosis, and induced systemic resistance (ISR) [26]. The result of previous research showed that *Pseudomonas spp* can protect plant of wheat against *Phytium spp*. and improve production of plant [20], induce peanut crops resistance from *Sclerotium rolfsii* pathogens [27], and *F. oxysporum* f.sp *fisi* & *Phytium ultimun* in peas [28]; can protect seedlings cucumber plants from *Colletotrichum orbiculare* [29].

It has been shown that *P. alcaligenes* KTS1, TRN2 and TMA1 suppress *Fusarium* disease in tomatoes [15]. Immerse tomato seeds with suspense *P. alcaligenes* deleted tomato diseaese by 45.44 -55.56% and could increase performance from 3 to 4 times than the control [30]. The cabbage (*Ipomoea reptans* Poir) immerses suspension treatments *P. alcaligenes* favored 25% germination, improving cultivation up to 24.4%, improved the number of leaves up to 23.15%, lengthening 25% of the stems, stretched the roots up to 46.90% and improved the roots fresh weight stems up to 67.07% and weight of the overy stem up to 84.21% than the control. Immerse the seeds with *P. alcaligenes* TRN2 treatment for 20 minutes showed best stimulated effect on seed germination [15].

Utilization of antagonistic microbes that live around plant roots like *Pseudomonas sp*., *Gliocladium sp*., *Bacillus spp*., and *Trichoderma spp*. is an environmentally friendly method controlling soilborne pathogens. Antagonistic microbes showed development of soilborne pathogens control [31]. *B. subtilis* inhibits the development of pathogens through the mechanisms of competition, antibiosis, and growth promotion. Suriani and Muis stated that *B. subtilis*, as one of the biological agents, also can promote plants growth and development [32]. The results of Wulansari's research showed that *B. subtilis* isolate can increased plant growth effectively, and *B. subtilis* B209 can increasing plant height by 2.78 cm/day that the highest result than other treatment [33]. Other research results show that *Bacillus sp*. strains BS 3, BS 4, and *Pseudomonas sp*. strains PF 1 and PF 3 can inhibit the development of *Fusarium oxysporum* in the form of clear zone that appears around the filter paper. The results showed that *Bacillus sp*. strain BS 3 and

strain BS 4 were Fungicidal [34]. Antibiotic type of *Pseudomonas sp*. strains PF 1 and PF 3 are fungistatic. Research by Pendango in 2018 showed that the soaking time of 15 minutes p with *Bacillus spp*. bacteria showed significant effect on amount of peanut seeds, namely 42.4 seeds, compared to the control with 35.2 seeds [35].

4. Conclusion

Pseudomonas alcaligenes (TmA1, TrN2, and KtS1) and *Bacillus sp*. (isolates 1,2,3, and 4) had different effects on the growth and yield of bitter melon. Mixing bacterial suspension on *P.alcaligenes*, *Bacillus sp*. isolates, and *P.alcaligenes* with *Bacillus sp*. isolate did not have a better impact on the yield and growth of bitter melon than the single treatment, but still better than the control. The bacterium *P. alcaligenes* TmA1, *Bacillus sp.* 2, and mixed *Bacillus sp.* gave the best effect on the weight of fresh bitter melon respectively, 89.25 g, 87.2 g, and 80.66 g compared to the control 66.32 g.

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