

## Efficiency of *Bacillus Cereus* Strain ATCC 14579 in Rice Washing Water Formulation as an Inoculant Agent in Seed Corn Growth

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**Abstract.** This study aimed to determine the efficient formulation dosage of rice washing water for *Bacillus cereus* Strain ATCC 14579 as an inoculant agent on seed growth of corn in a micro plotscale. The experiment method was used by randomized block design factorial, consisting of two factor, the first factor is variety, namely V1: Anoman variety and V2: Bima URI 19 type, the second factor is dosage of inoculant agent in rice washing water formulation, namely D1= 20 mL inoculant agent/kg of seed, D2= 40 mL inoculant agent/ kg of seed, D3= 60 mL inoculant agent/ kg of seed, D4= 80 mL inoculant agent/kg of seed, D5= 100 mL inoculant agent/kg of grain and D6= control (without formulation dosage). Each treatment was repeated three times, so that there were 42 experimental units. The result showed that the formulation dosage of 80- 100 mL inoculant agent/kg of seed efficiently increased the growth of seed corn varieties Anoman and Bima URI 19.

**Keywords:** *Bacillus cereus*; organic waste; carrier media

### 1. Introduction

Using rhizobacteria as biological agents is appropriate because it is more environmentally friendly. Biological control using microorganisms associated with the rhizosphere and organic matter is an efficient and environmentally friendly approach [1]. Besides, some rhizobacteria can provide, mobilize, and facilitate the absorption of various nutrients in the soil because of their ability to bind free nitrogen and produce growth hormones. Some rhizobacteria make plant growth-stimulating hormones, increasing the dry weight of maize plants [2].

One of the rhizobacterial groups that have been widely studied is *Bacillus* sp. Its rapid ability to colonize plant roots and extensive environmental adaptation because *Bacillus* scattered in nature, especially in the rhizosphere. *Bacillus* sp. can be found in soil, water, air, and decomposed plant residue [3]. *Bacillus* exists in the ground. It is active at the soil surface because it utilizes organic matter and root exudates. *Bacillus* sp. has proven ideal for development as a stable and efficient biological product because it produces heat-resistant endospores [4].

*Bacillus cereus* is a species of *Bacillus*, widely reported as a biofertilizer, bioremediation, and bioprotectant agent. To enhance plant growth, this species is directly involved in increasing nitrogen uptake, phytohormone synthesis, dissolving minerals such as phosphorus, and the secretion of siderophore, which chelates iron and makes it available to plant roots [5]. *Bacillus cereus*' role as an efficient phosphate solvent and biopesticide against fungal pathogens has been documented previously [6]. Likewise, these bacteria's ability to synthesize IAA and increase growth has been proven [7].

By looking at the size of *Bacillus cereus*'s role in plants' growth and production, it is necessary to multiply these bacteria to help increase plant productivity. Rhizobacteria medium using synthetic is limited to laboratory scale. Moreover, for a larger scale, it is expensive when using synthetic medium. Therefore, it is advisable to look for an organic medium that is cheap and easy to obtain [8]. Some organic Wastes that can be used as a medium for bacterial propagation are coconut water waste [9], liquid tofu waste [10], soy washing water, and rice washing water [11]. Coconut water waste contains glucose, sucrose, inositol, fructose, sorbitol, protein, minerals, vitamins B and C [12]. Rice washing water (leri water) contains N, P, K, and C, vitamins B1 and B12. The compound contained in the liquid

waste is needed for bacterial growth. The selection criteria for medium propagation should include cost, availability, chemical stability, toxicity levels, and farmer convenience to management and flexibility [13]. This study aimed to determine the efficient formulation dosage of rice washing water for *Bacillus cereus* Strain ATTC 14579 as an inoculant agent on Anoman and Bima URI 19 seed corn varieties.

## 2. Research Method

The research was conducted at the plant pest and disease laboratory, Maro's Cereal Research Center. Then, proceed with a micro plot scale application in a greenhouse at the exact location. The Materials used include isolates of *Bacillus cereus* strain ATCC 14579, Nutrient Agar medium, rice washing water, coconut water, molasses, tryptic soy broth (TSB), sterile water, aluminum voile, wrapping, Alcohol 70 %, methylated spirits, and HVS paper. The tools used include Petri dishes, Erlenmeyer, stirrer, scale, hotplate, pipette, autoclave, Bunsen, measuring cup, ose needle, shaker, oven, and laminar airflow and spectrophotometer. The experiment used a factorial randomized block design in the two-factor with three replications. The first factor is a variety of seed corn: V1: Anoman, V2 = Bima 19 URI. The second factor is dosage of inoculant agent in rice dishwater formulation, namely: D1 = 20 mL inoculant agent/kg of seeds, D2 = 40 mL inoculant agent/kg of seeds, D3 = 60 mL inoculant agent/kg of seeds, D4 = 80 mL inoculant agent/kg of seeds, D5 = 100 mL inoculant agent/kg of seeds and D6 = control.

Cultivation *Bacillus cereus* was cultivated on Nutrient Agar (NA) media in a petri dish, then incubated at room temperature for 2 x 24 hours. Then, the bacteria are harvested in sterile distilled water, ready to be used as a blow. Provision of formulation medium, rice washing water is used as media for propagation. The organic waste is separately put in 50 mL of 250 mL Erlenmeyer, then added with Tryptic Soy Broth (TSB) 10%, then distilled water to reach a volume of 200 mL. The media mixture is sterilized in an autoclave at 121OC for 15 minutes; after sterilization, the media is cooled and ready for use. Preparation of Planting Media: The planting medium used is sterilized soil+sand (1:1), then put in a polybag as much as 9 kg/polybag. Application Micro plot inoculants, the corn seeds before planting, were inoculated with organic media according to the previous treatment dose. When inoculating the seeds, enough Arabic gum is added as an adhesive; they are air-dried for 2 hours, and the seeds are ready for planting. One seed per hole is planted, and ten seeds per polybag. At the age of two weeks of planting, thinning is done, leaving three plants. Research variables: Plant height, root length, and fresh weight were measured by taking five samples 28 days after planting.

## 3. Result and Discussion

Based on observations of plant height average, it can be seen that the use of the seed corn varieties Anoman and Bima URI 19 has a significant effect on plant height at the 0.05 level. The LSD test results showed that types gave the plant height average for Anoman varieties, namely 43.52 cm, but tended not to be different from the Bima URI 19 variety, which was 38.46 cm (Table 4). At the same time, the inoculant dosetreatment and its interactions have no significant effect.

**Table 1.** The average plant height (cm) 28 days after planting

Treatment	V1	V2	Average
D1	36.27	38.46	37.36
D2	43.13	37.70	40.41
D3	46.20	36.23	41.21
D4	43,27	43.73	43.73
D5	44.87	41.47	43.17
D6	47.13	33.20	40.16
Average	43.52 <sup>a</sup>	38.46 <sup>a</sup>	
<i>LSD</i> 0.05	8.459		

Note: A number followed by the same letter (a) means no different

The use of *Bacillus cereus* strain ATCC 14579 as an inoculant agent in rice washing water formulation, and its interactions have no significant effect on plant height. The highest plant height average was found on treatment D4, 43.73 cm, and the lowest on treatment D1, 37.36 cm (Figure 1).

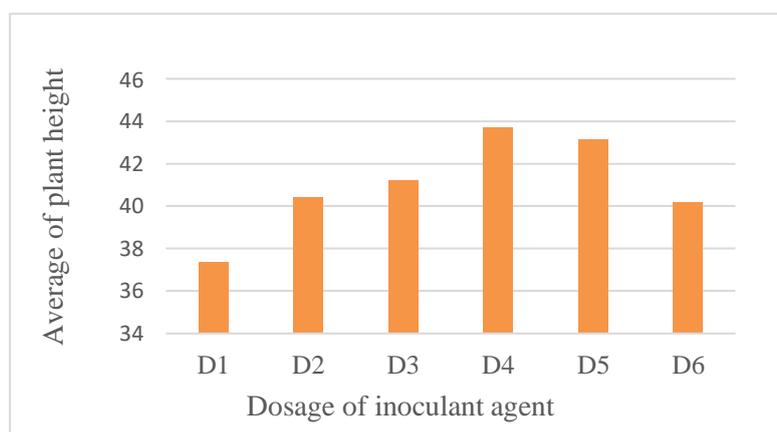


Figure. 1. The average plant height (cm) 28 days after planting

Based on the observation of root length, the formulation dose and variety of seed corn significantly affect the 0.05 level. *LSD* test results (Table 2) show that the treatment D5: inoculant dose of 100 ml/1 kg of seeds gave the highest root length, namely 9.10 cm, tending to be the same as D3: inoculant dose of 60 ml/1 kilograms of sources, but distinct from other dosages.

**Table 1.** The average plant height (cm) 28 days after planting

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Based on observing plants' wet weight average, the formulation dose and variety of seed corn do not significantly affect the 0.05 level. The highest plant wet weight average was found on treatment D3: inoculant dose of 60 ml / 1 kg of seeds, namely: 28.2 gr cm Anoman variety, and the lowest was found on treatment D6: control, namely: 9.2 gr Bima URI 19 variety (figure 2).

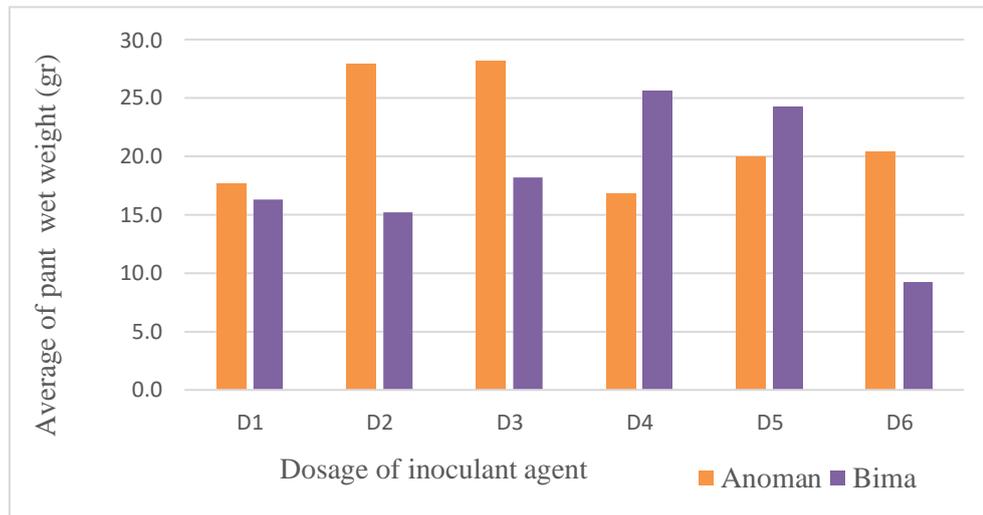


Figure. 2. The average root length (cm) 28 days after planting

The result was that the Anoman variety responded more to all parameters. Differences in characteristics, especially genetics, can cause it. Genetic factors in plants are one of the reasons for the differences in response between plants because of differences in genes that regulate these characteristics. Each plant shows various growth and yields due to genetic and environmental influences, where genetic influence is the effect of heredity possessed by each strain or variety. In contrast, ecological influence is generated by habitat and environmental conditions [14].

Isolate of *Bacillus cereus* strain ATCC 15579 as an inoculant agent on the seed gives a good to plant height and root length parameters. Rhizosphere is a zone of intense microbial activity with the growth of plants. Many beneficial microorganisms of agricultural importance are rhizosphere colonizing species that can increase plant growth via various mechanisms [15]. Microbial inoculants applied as seed treatments deliver microorganisms directly to the plant rhizosphere, the narrow soil zone surrounding the roots where plants interact directly with microorganisms [16]. Similarly, inoculating seeds with a wide range of putative and endophytic bacterial isolates improved shoot dry weights in maize seedlings [17]. The micro-scale plot test for growth factors showed that the formulation dose of 80-100 ml inoculant agent/ 1 kg of seed efficiently increased plant height, root length, and fresh weight of corn varieties Anoman and Bima 19 URI 28 days after planting. However, waiting for the research results on a field scale is still necessary to ensure this dose is recommended.

#### 4. Conclusions

*Bacillus cereus* strain ATCC 14579 can be used as an inoculant agent on rice washing water formulation. The formulation dosage of 80-100 ml inoculant agent/ 1 kg of seed is an efficient dose that can increase the growth factor of corn varieties Anoman and Bima URI 19.

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