Comparative evaluation of cocoa seeds germination in local variety Gantarangkeke Bantaeng (GTB) with MCC 01 variety using seed priming treatment

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Abstract. Gantarangkeke Bantaeng (GTB) variety seed is a type of local seed that has experienced a decline in its existence due to the emergence of superior seed types such as the MCC 01 variety resulting in low local cocoa production due to the lack of use of GTB seeds in South Sulawesi. One of the causes of low seeds capability is the lack of special treatment given to seeds which refers to the inhibition of germination of cultivated cocoa seeds. Seed priming is a treatment of soaking seeds with microbes to improve the quality of seeds germination. This study aims to compare the germination of local varieties of cocoa seeds GTB with varieties MCC 01 using Plant Growth Promoting Rhizobacteria (PGPR) seed priming treatment. This study employed a randomized block design (RBD). Two cocoa seed varieties used as the first factor consisted of local GTB and MCC 01, while the seed priming treatment as the second factor consisted of PGPR concentrations of 0% (control), 5%, 10%, and 15%. The results indicate that the seed priming treatment affected the germination of two cocoa varieties seeds which included the parameters of MCC 01 seeds germination and the growth speed which was higher than local GTB seeds, 100.00% and 7.14%/etmal, respectively. Abnormal seedlings in local GTB variety were higher than MCC 01 seeds (10.00%). It can be concluded that the seed priming treatment of cocoa seeds has a high effect on seed germination of the MCC 01 variety compared to the local variety GTB.

1. Introduction
The soaring increase in world cocoa demand is due to the increasing demand for cocoa beans which is getting higher every year. The annual world demand for cocoa reaches 6.7 million tons and only 2.5 million tons can be met. This means that there are still more than 4 million tons to meet the increasing market needs, so that this can still be an opportunity for Indonesia, especially South Sulawesi [1].

Cocoa is a strategic commodity which ideally is able to play a maximum role in increasing the income of farmers in South Sulawesi, especially for farmers in Bantaeng Regency. This can happen because cocoa has always had a good price development in meeting market needs. But in reality, although every year there is an increase in the area of planting, it does not necessarily increase production. The average farmer production is only 1/5 of the potential productivity of the plant, which ranges from 1.2 - 3 tonnes per hectare [2].

South Sulawesi is one of the largest cocoa contributors in Indonesia. Various new superior cocoa clone seeds have been created and disseminated to maximize the role of South Sulawesi in supporting
cocoa needs in Indonesia. In addition, local seeds have also become one of the most defended seeds in South Sulawesi, one of which is the Gantarangkeke Bantaeng (GTB) clone seed. GTB local seed is one type of local seed that has decreased its existence due to the emergence of superior seed types such as the MCC 01 variety, resulting in low local cocoa production due to the lack of use of GTB seeds in South Sulawesi. One of the causes of low seeds capability is the lack of special treatment given to seeds which refers to the inhibition of germination of cultivated cocoa seeds. On average, farmer production is only 1/5 of the potential productivity of the plant, which ranges from 1.2 - 3 tonnes / Ha in South Sulawesi.

This statement indicates the emergence of a problem currently being faced, namely whether to increase cocoa production to meet cocoa needs in the future, while the availability of good capability seeds is currently difficult to obtain. Cultivation of cocoa starts from the nursery stage, namely the process of seed germination and from this stage it will determine the success of the plant to grow well in the future. In this nursery period, good quality seeds must be obtained so that it will be the benchmark for optimum growth which is marked by good quality cocoa beans. Of course, in order to obtain quality seeds, the selection or use of good seeds in germination will be a major factor. One way to get good seeds is by curing or priming the seeds. Priming is a hydration activity slowly before the seeds are germinated so that the potential for seed water can reach a balance to activate metabolic activities in the seeds to improve germination and seed growth [3].

Priming treatment can be combined with the provision of biological agents capable of improving the capability of seed germination, for example with microbes that are capable of producing growth hormones in plants. This treatment is known as bio-priming. One of the microbes that can be utilized are microbes that are included in the Plant Growth Promoting Rhizobacteria (PGPR) group which can produce plant hormones and can help provide more nutrients to spur plant growth.

In the rhizosphere or bamboo roots there are *Pseudomonas fluorescens* and *Bacillus polymixa* bacteria which can help the decomposition process (decomposer). The bamboo root PGPR bacteria can secrete a liquid that is able to dissolve minerals so that they become available nutrients, break down and decompose organic matter (decomposition of organic matter) into plant nutrients. In addition, the *Pseudomonas fluorescens* and *Bacillus polymixa* bacteria can release enzymes and substances that are useful to stimulate plant growth and release antibiotics that can inhibit plant growth and release antibiotics that can inhibit the growth and development of pathogenic microbes (microbes that cause disease) [4].

This study aims to obtain data and information about the Comparative Evaluation of Cocoa Seeds Germination in Local Variety Gantarangkeke Bantaeng (GTB) with MCC 01 Variety Using Seed Priming Treatment. Information from the results of this study is expected to be a solution for cocoa farmers in improving their cultivation systems, especially in the nursery phase and selecting good seeds.

2. Methods

2.1. Location, time and materials

The research was carried out in the Talaka Farmers Group Garden, Gantarangkeke District, Bantaeng Regency, South Sulawesi, Indonesia. This research was carried out from August to November 2019. The equipment used were a 30 liter bucket, hoe, shovel, scissors, knife, machete, ruler, meter, callipers, pH meter, wheelbarrow, analytical balance, sprayer, scale, jerry can, funnel plastic aqua bottles, small hoses, stoves, pans, paranets, cloths, plastic cups, brown envelopes, millimeter-block paper, cameras, nameplates, lighters, and stationery. The materials used were soil samples from the study site, local GTB (Gantarangkeke Bantaeng) cocoa seeds, MCC 01 cocoa seeds, bamboo rhizosphere (*Bambusa blumeana*), molasses, bran, raw shrimp paste, rice water, water, husk charcoal, sawdust, clear plastic, dab soap, 12 x 17 cm polybags, label paper, dolomite and label paper.

2.2. Methods and data analysis

The study was conducted using a two-factor factorial design (F2F) in a Randomized Block Design (RBD) as an environmental design. This research consisted of 2 factors. The first factor is the type of
cocoa seedling consisting of: local cacao seed GTB (Gantarangkeke Bantaeng) and cocoa seed clone MCC 01, while for the second factor, the bio-priming treatment using bamboo rhizosphere PGPR solutions on cocoa seeds with various concentrations consisted of 0% concentration (0 ml PGPR solution or 1000 ml water), 5% concentration (50 ml PGPR solution + 950 ml water), 10% concentration (100 ml PGPR solution + 900 ml of water) and a concentration of 15% (150 ml of PGPR solution + 850 ml of water). The implementation of the method in this research involved making a plant growth promoting rhizobacteria solution which was conducted by taking the bamboo rhizosphere and then grown by fermentation in the container for 14 days, seed preparation which are 80 seeds of GTB and 80 seeds of MCC 01 by selecting seeds from MCC 01 and GTB cocoa pods and then separating the placenta in the seeds, application of the PGPR solution to cocoa seeds (priming cocoa seeds) together according to the treatment for 18 hours [5], preparation of sprouts media by using sterile cloths and sawdust, planting in seedbed (nursery), maintenance and observations.

2.3. Observation parameters
Observation parameters in this study are as follows:

2.3.1. Germination capacity (\%)
Measurement was carried out by observing the number of normal germinated seeds marked with cotyledons on the raised seed. Calculation of germination that is on day 7 and day 14 [6]. The parameter measured the comparative germination capacity of cocoa seeds germination of the local variety GTB and MCC 01 variety. Sprouts are calculated using the equation (1):

$$ DB = \frac{\sum KN \text{ Observation I} + \sum KN \text{ Observation II}}{\text{Number of Seedlings Planted}} \times 100\% $$

Information:
\( DB = \) Germination Capacity (\%)
\( \sum KN \text{ Observation I} = \) The number of sprouts is normal on the 7th day
\( \sum KN \text{ Observation II} = \) The number of sprouts is normal on the 14th day

2.3.2. Seed growth speed (\%/etmal)
Measurement was based on observing the number of normal germinating seeds every day until the day 14 and expressed on a percent scale [7]. This parameter measures comparative seeds growth speed of cocoa seeds germination in local variety GTB and MCC 01 variety. Seed growth rate is calculated using the equation (2):

$$ KCT = \left( \% \frac{KN}{etmal} \right) = \sum_{t=0}^{tn} \frac{N}{t} $$

Information:
\( t = \) Observation time
\( N = \) Percentage of normal sprouts every time of observation
\( tn = \) end time of observation (day 14)
\( etmal = 1 \text{ day (24 hours)} \)

2.3.3. Abnormal seeds (\%)
Measurement was based on observing the number of seeds that germinate abnormally. Observation of abnormal seeds is carried out on the day 14 after the nursery [6]. This parameter measures comparative abnormal seeds of cocoa seeds germination in local variety GTB and MCC 01 variety. Abnormal seeds are calculated using the equation (3):

$$ \text{Abnormal Seeds} = \frac{\sum \text{Abnormal Seeds}}{\text{Number of Seedlings Planted}} \times 100\% $$
3. Results and discussion

The results show that the comparison of the two types of cocoa seeds in the bamboo rhizosphere PGPR priming treatment with the seed concentration and the interaction of PGPR application. This comparison had sufficiently effect on seed germination, seed growth speed and abnormal seeds of cocoa.

3.1. Germination capacity

The observations showed that the comparison of the two types of cocoa seeds (GTB and MCC 01) in the treatment of bamboo rhizosphere PGPR application with the concentration of seed priming and the interaction of PGPR application on the two types of cocoa seeds had sufficiently effect on the germination capacity of cocoa seeds. The average germination power is presented in (figure 1).

![Average Germinations Capacity](image1)

**Figure 1.** Average seed germination capacity of two cocoa clones as affected by PGPR concentration in seed priming.

3.2. Seeds growth speed

The observations showed that the comparison of the two types of cocoa seeds (GTB and MCC 01) in the seed priming treatment of bamboo rhizosphere PGPR with seed immersion concentration and the interaction of PGPR application on the two types of cocoa seeds sufficiently affected the speed of growing cocoa seeds. The average seed growth rate is presented in (figure 2).

![Average Seeds Growth Speed](image2)

**Figure 2.** Average seed growth speed of two cocoa clones as affected by PGPR concentration in seed priming.
3.3. Abnormal seeds
The observations showed that the comparison of the two types of cocoa seeds in the seed priming treatment of bamboo rhizosphere PGPR with seed immersion concentration and the interaction of PGPR application on the two types of cocoa seeds sufficiently affected the abnormal seeds of cocoa. Average abnormal seeds are presented in (Figure 3).

![Average Abnormal Seeds](image)

**Figure 3.** Average percentage of abnormal sprouts of the two cocoa clones as affected by PGPR concentration in seed priming.

3.4. General discussion

3.4.1. Comparison of the seeds of MCC 01 with GTB. In seed germination in the nursery phase, the results showed that the ratio of MCC 01 cacao seed types had the highest effect on germination and seed growth speed, while local GTB seeds had the highest effect on abnormal seed conditions. The seeds of MCC 01 had sufficiently effect on germination, which was counted twice on the 7th and 14th day with the highest germination rate of 100%, the observed seed growth rate for 14 days with the fastest seed growth percentage of 7.14%. In total and the percentage of abnormal seeds calculated on the 14th day with the highest abnormal seeds, namely 10%. This is due to the influence of the germination environment where the seeds grow at the time of the nursery, where the humid conditions and the long seeding time lead to optimum germination and seed growth speed. In addition, the genetic factor of the seed itself is also a major influence on germination, because if the genetic of the seed is not strong enough to accept the environmental conditions in which it is grown and the initial treatment, it will cause the seeds to not have the potential to germinate normally or in other words, abnormal seeds. The process of seed germination is influenced by genetic and environmental factors. The improvement of the external environment will significantly encourage the emergence of radicles as the beginning of the seed germination process. The emergence of sprouts above the soil surface is a factor that reflects the vigor of a seedling. To find out which treatment can increase vigor, the sprouts that can appear above the soil surface are observed from the number of seeds germinated.

The average germination rate with the highest effect was the type of MCC 01 seed when compared to the local GTB seed germination capacity. The average seed germination rate in this study had optimum growth because the seeds had been given previous treatment, where the highest germination growth in MCC 01 seeds and local GTB seeds had been treated with PGPR with a concentration of 5% and 15%. In accordance with the statement of the Jambi Agricultural Training Agency (2010), which states that the PGPR content is dominated by *Pseudomonas fluorescens* and *Bacillus polymixa*. The benefit claims obtained are as a bioprotectant, biofertilizer and as a biostimulant. It is also in line with the research of [8], which states that several bacterial general obtained from the rhizosphere of bamboo
plants include: genus *Bacillus*, *Pseudomonas*, *Enterobacter*. So that it absolutely giving more influence to different condition in both MCC 01 and GTB seeds on germination process. Application of PGPR treatment with the highest concentration is a major factor in the germination power of seeds. This is in line with the opinion expressed by [9], that pre-treatment is carried out with the aim of increasing the power, speed and uniformity of seed germination.

The average seed growth speed with the fastest effect was the MCC 01 seed type when compared to the local GTB seed growth rate. The average seed growth speed of MCC 01 seeds and local GTB seeds with PGPR treatment with a concentration of 5% and 15% showed the fastest growing results when compared to other seeds. The growth rate of MCC 01 seeds and local GTB seeds is influenced by the optimum condition of the seeds at the time of the seed nursery and the provision of special treatment to the seeds so that the metabolism of seed growth can run well. This is in line with the statement of [10] stated that the speed of seed growth is a process of rapid seed reactivation if the surrounding conditions are for optimum growth and the metabolic process is not hampered.

Abnormal seeds in the use of local GTB seeds without PGPR treatment or control and with PGPR treatment with a concentration of 5% showed the highest yield of abnormal seed conditions when compared to other seeds. Abnormal seeds occur due to the lack of pre-treatment of local GTB seeds, besides the physical and genetic characteristics of local GTB seeds which tend to be weaker than MCC 01 seeds. The occurrence of abnormal seeds in the nursery process is due to the deterioration of the seeds so that the quality of the seeds decreases which resulting in inhibition of the process Germination.

3.4.2. Interaction of the concentration of PGPR priming seeds and the use of two types of cocoa. In seed germination, the interaction between the use of two types of cocoa with seed priming concentration treatment with PGPR had a very significant effect on germination, germination speed and the percentage of abnormal seeds. This was because the cells in the MCC 01 cocoa seed and the local GTB seed after being given priming treatment with PGPR became more active in influencing seed metabolism. This is in line with the statement of [11], which states that direct seed priming is a technique for seed invigoration through controlled water imbibition. Currently, invigoration is an alternative that can be used to overcome low quality seed quality by treating seeds before planting by reactivating seed metabolism so that the seeds are ready to enter the germination phase. In addition to the invigoration process, the immersion process results in simultaneous germination and reduces environmental stress. The synchronization of the appearance of the radicle will affect the uniformity of the length of the hypocotyl, this parameter will then show the variety of seeds in the ability to lift the cotyledons.

4. Conclusion
The comparison of MCC 01 seed growth with local GTB seeds through seed priming treatment gave different responses to cocoa seeds germination including germination parameters with an optimal value of 100.00%, seed growth rate with an optimal value of 7.14%/etmal, and abnormal beans with a value optimal 10.00%. So that the seeds priming treatment with PGPR has an effectively effect on the germination of two types of cocoa seeds. This comparison indicates that MCC 01 seeds are optimal for use in cocoa seeds germination, which require a shorter time with most of sufficiently great effect rather than local GTB seeds especially the evaluation of seeds priming treatment. Based on the results of this study, it is hoped that in the future, cocoa farmers can use the seed priming method with the bamboo rhizosphere PGPR combined with the MCC 01 South Sulawesi clone seeds to obtain maximum cocoa seed germination results.

References

[1] Yusniar 2013 *Membangun Kesejahteraan Petani Lewat Nagari Model Kakao (NMK)* (Padang: Dinas Perkebunan Sumatera Barat Indonesia)

[2] Junaedi J, Thamrin S and Baha B 2017 Kajian penggunaan klon unggul kakao pada perkebunan rakyat di Kabupaten Bone Agroplantae: Jurnal Ilmiah Terapan Budidaya dan Pengelolaan Tanaman Pertanian dan Perkebunan 6 (2) 46-49.
[3] Rouhi H R, Surki A A, Sharif-Zadeh F, Afshari R T, Aboutalebian M A and Ahmadvand G 2011 Study of different priming treatments on germination traits of soybean seed lots Notulae Sci Biol. 3 (1) 101-108

[4] Effendi M S 2011 Kinetika fermentasi asam asetat (vinegar) oleh bakteri Acetobacter aceti dari etanol hasil fermentasi limbah cair pulp kakao Jurnal Teknologi dan Industri Pangan 8 (2) 125-135

[5] Ratnawati, Saputra S I and Yoseva S 2013 Waktu Perendaman Benih dengan Air Kelapa Muda Terhadap Pertumbuhan Bibit Kakao (Theobroma cacao L.) (Pekanbaru: Fakultas Pertanian Universitas Riau Indonesia)

[6] Debtisari H E, Erawati D N and Sugiyarto 2018 Pengaruh cara penyimpanan terhadap viabilitas benih kakao (Theobroma cacao L.) Klon Sulawesi 01 Agropross National Conference Proceedings of Agriculture (Jember: Politeknik Negeri Jember Indonesia)

[7] Tefa A 2017 Uji viabilitas dan vigor benih padi (Oryza sativa L.) selama penyimpanan pada tingkat kadar air yang berbeda Jurnal Pertanian Konservasi Lahan Kering 2 (3) 48-50

[8] Susanti I, Retno W K and Fatim I 2007 Uji Sifat Probiotik Bakteri Asam Laktat Sebagai Kandidat Bacteria (Spain: Springer International Publishing)

[9] Schmidt L 2002 Guidelines for Handling Tropical and Sub-Tropical Forest Seed Plants (Humlebaek: Danida Forest Seed Centre)

[10] Leisolo M K, Riry J and Matatula E A 2013 Pengujuan viabilitas dan vigor benih beberapa jenis tanaman yang beredar di pasaran Kota Ambon Jurnal Agrologia 2 (1) 1-9

[11] Heydecker W 1973 Germination of an Idea: The Priming of Seeds (Nottingham: School of Agriculture Research, University of Nottingham UK)