Bio-priming of Aceh local chili seeds in the effort to increase production and begomo virus resistance

S Hafsah1*, Yusnizar2, Nura1, K S Kaloko3, F Reza1, Firdaus4
1Department of Agrotechnology, Faculty of Agriculture, Universitas Syiah Kuala, Jalan Tgk. Hasan Krueng Kalee 3, Darussalam-Banda Aceh, 23111, Indonesia
2Department of Soil Science, Faculty of Agriculture, Universitas Syiah Kuala, Jalan Tgk. Hasan Krueng Kalee 3, Darussalam-Banda Aceh, 23111, Indonesia
3Bachelor Program of Agrotechnology Department of Agrotechnology, Faculty of Agriculture, Universitas Syiah Kuala, Jalan Tgk. Hasan Krueng Kalee 3, Darussalam-Banda Aceh, 23111, Indonesia
4Assessment Institute of Agricultural Technology of Aceh, Indonesia

*Email: sitihafsah@unsyiah.ac.id

Abstract The purpose of this study was to determine whether P60 could induce resistance in local Aceh chili varieties to begomovirus and compared with national varieties. This research was conducted at the Experimental Farm, Genetics and Plant Breeding Laboratory, Faculty of Agriculture, Syiah Kuala University, Banda Aceh, from March to July 2021. This study used a factorial randomized block design. The tested factors consisted of two factors. The first factor is bio priming. The second factor consists of four local Aceh chili varieties, namely: V1 = Odeng, V2 = Lanyoe, V3 = Super Lamando and V4 = LamandoLapaben and one national variety, namely V5 = Baja F1 as a comparison variety. The results showed that the best local Aceh variety was LamandoLapaben for the variables of resistance response, incubation period, disease intensity, disease incidence, stem diameter, and crown width. The best local variety Aceh Lanyoe on plant height and dichotomous height parameters. Giving P60 can reduce the percentage of disease incidence by 52.00% and slow down the process of emergence of disease symptoms (incubation period) 27.11 DAP. The best combination treatment was the LamandoLapaben variety, which was 1.41 milligram Units-1 (Umg)-1.

1. Introduction
Chili (Capsicum annuum L.) is one of the leading horticultural crop commodities which is widely cultivated because of its economic value and nutritional content. Chili is used in a mixture of food, medicine and some species are used as ornamental plants [1] According to [2] the demand for chili is increasing along with the increase in population. Chili planting in the dry season is generally more disturbed by insect pests. One of the big losses in chili farming is that chili plants are attacked by insect pests such as whitefly (Bemisia tabaci Genn) which is the vector of the virus [3] The epidemic of diseases caused by Begomovirus is increasingly widespread and has the potential to inhibit the production of chili plants, so it is necessary to have a procedure to detect Begomovirus in plants, including serological methods and PCR Polymerase chain reaction (PCR).
One of the efforts to increase the yield of chili and increase resistance to various viruses is to use quality seeds of a variety [4]. Chili basically consists of hybrid and non-hybrid (local) varieties, each of which has several advantages and disadvantages. Bener Meriah Regency is one of the areas that has several genotypes of local Aceh chili that have been cultivated well by farmers. The dominant genotypes planted by farmers are chilies Odeng, Lanyoe, Super Amando, and Lamando Lapaben which have long been known by farmers because they have superior productivity with good fruit quality. [5] stated that the results of the characterization of four local varieties of Aceh, Odeng and Lamando Lapaben had higher productive branches than Lanyoe and Super Amando.

Priming is to slowly hydrate the germinated seeds, to activate T. harzianum metabolism in seeds [6] and [7] stated that soybean seeds coated with 2 percent plus P60 could increase the vigor index value and seed germination. [8] added that the bio-invigoration of seeds using Bacillus sp. CKD061 was able to increase viability and seed vigour of local upland rice cultivars. The treatment showed that the seed vigour index increased by 133% in the Waburi-buri cultivar and 127% in the Daindo Mornene cultivar when compared to the control [9].

Increased productivity of chili can be done by improving genetic potential through the formation of hybrid varieties and the use of superior varieties [10]. The use of bio-priming on local varieties of Aceh will increase germination vigour, growth and will maximize yield potential and resistance to Begomovirus.

2. Materials and methods

2.1. Location and period
The location where the study conducted was the Experimental Field, Genetic and Plant Breeding Laboratory of the Faculty of Agriculture, Universitas Syiah Kuala, Banda Aceh, Indonesia during the period of March to July 2021.

2.2. Supplies and equipment
Supplies and equipment used during the experiment were plastic mulch, meters as measure, hoes for land preparation, seedling tray and other equipment such as dipper, hand sprayer and stationaries. The materials used were chili seeds of Lanyoe, Super Lamando, Lamando Lapaben a local variety from Aceh, AB Mix fertilizer and Pseudomonas Flurorescdens P60 bacteria.

2.3. Field culture, planting, and harvesting
Land preparation is carried out one month before planting the seeds with the aim of loosening the soil, namely through plowing the soil and applying manure. The beds are made with a length of 2.5 m and a width of 1 m. The distance between the experimental plots is 0.5 m while the spacing is 50 cm x 50 cm. The beds were covered with silver black plastic mulch (MPHP). P60 prepared and prepared for seed treatment. The P60 used is P60 bio bacteria that has a volume of 1000 ml. Then the local chili seeds were soaked in P60 suspension with water for approximately 30 minutes with a ratio of 1:1. Chili seeds that have been treated with P60 and control, then planted in a seed tray filled with sterile media (soil+compost+husk charcoal) each with a ratio of 2 : 1 : 1).

2.4. Experimental design and analysis
This study used a factorial randomized block design. The tested factors are 2 levels of treatment (P0= non P60, P1 = add P60). The second factor is Aceh’s local chili varieties, namely: V1 = Udeng, V2 = Lanyoe, V3 = Super Lamando and V4 = Lamando Kapaben and V5 = Baja F1. So, we get 2 x 5 = 10 combinations. Each treatment was repeated 3 times to obtain 30 experimental units. Each experimental unit consisted of 10 sample plants so that the total plant population was 300 plants. The transfer of chili seedlings to the field is carried out five weeks after sowing (MSS). Plant maintenance includes watering 2 times a day, weeding once a week, fertilizing, and controlling pests and diseases.
Fertilization of chili plants using AB mix. Fertilizer A and fertilizer B are diluted with sufficient dissolution of fertilizer to 5 L of water. Watered on plants around the roots as much as 200 ml per plant.

2.5. Observation parameters
The observed variables were the incubation period and disease incidence, plant height, dichotomous height, stem diameter, crown width and peroxidase enzyme analysis.

3. Results and discussion

3.1. Incubation time and disease occurrence
Table 1 showed the average incubation period of the tested varieties, which ranged from (16.79 – 33.79 days). The average incidence of disease in the varieties tested was in the range (51.94 – 61.94 %). The Lamando Lapaben variety gave the longest symptoms of begomovirus attack (26.29 Days) which was statistically significantly different from the comparison variety Baja F1 (33.79 Days).

In the parameter of disease incidence, the Lamando Lapaben variety showed the lowest disease incidence (55.00%) which was statistically significantly different from the comparison variety Baja F1 (51.94%). In this study, the resistance character between local Aceh chili varieties differed in showing the resistance response to begomovirus. [11] states the resistance character of each genotype to different pests or diseases resulting in a different number of plant deaths for each genotype. [12] states that the nature of resistance is controlled by genes in the nucleus (nuclear genes).

| Genotype            | Resistance Response | Incubation Period (Days) | Disease Intensity (%) |
|---------------------|---------------------|--------------------------|-----------------------|
| Odeng               | 19,98 ab            | 61,94 b                  |
| Lanyoe              | 21,45 ab            | 60,55 b                  |
| S. Lamando          | 16,79 a             | 55,83 ab                 |
| Lamando Lapaben     | 26,29 ab            | 55,00 ab                 |
| Baja F1             | 33,79 b             | 51,94 a                  |
| BNJ 0.05            | 15,80               | 8,13                     |

3.2. P60 treatment

| P60 Treatment | Incubation Period (Days) | Disease Intensity (%) |
|---------------|--------------------------|-----------------------|
| P1            | 27,11 a                  | 52,00 a               |
| P0            | 16,63 b                  | 62,11 b               |
| BNJ 0.05      | 7,4                      | 3,81                  |

Note: Numbers Followed by the Same Letter in the Same Column Are Not Significantly Different Based on the Honestly Significant Difference Test (BNJ) level 0.05,

The effect of giving P60 showed the lowest incidence of disease (52.00%) which was statistically significantly different from without giving P60 (62.11%). This is consistent with research by [13] which stated that P60 antagonist bacteria have the ability to colonize plant roots after being inoculated into seeds and induce plant resistance. The results of research from [14] P60 bacteria have Plant Growth Promoting Rizobacteria (PGPR) properties that produce antibiotics that can inhibit the growth of pathogens.
In chili plants that are susceptible to Begomovirus infection when attacked early in growth will cause fruit size to be short. Begomovirus infection that occurs in the late growth phase does not significantly affect yield reduction, but if Begomovirus attack occurs in the vegetative phase it will reduce yields significantly [15].

3.2. Plant height and dichotomous height
The average high crops and local varieties of chili dichotomous in Aceh due to the granting of P60 is shown in Table 2. The average height of the local varieties tested ranged between 46.74 to 57.33 cm. In the average parameter of dichotomous height that was tested, which ranged from 18.72-28.90 cm. The Lanyoe variety has the highest plant height (57.33 cm) which is statistically significantly different from the comparative Baja F1 variety (55.65 cm). The Lanyoe variety has the highest dichotomous height (26.80 cm) which is statistically significantly different from the comparative Baja F1 variety (28.90 cm).

Table 2. Average value plant height and dichotomous height in Aceh local varieties due to P60 treatment.

| Genotype       | Vegetative Growth |          |          |
|----------------|-------------------|----------|----------|
|                | Plant Height (cm) | Dichotomous Height (cm) |          |
| Odeng          | 46.74 a           | 18.72 a  |          |
| Lanyoe         | 57.33 b           | 26.80 ab |          |
| S. Lamando     | 54.74 ab          | 22.72 ab |          |
| LamandoLapaben | 53.06 ab          | 21.21 ab |          |
| Baja F1        | 55.65 ab          | 28.90 b  |          |

| BNJ 0.05       | 9.34              | 9.40     |          |

P60 Treatment | Plant Height (cm) | Dichotomous Height (cm) |
|--------------|-------------------|-------------------------|
| P1           | 54.61             | 24.38                   |
| P0           | 52.40             | 22.96                   |

BNJ 0.05

Note: Numbers Followed by the Same Letter in the Same Column Are Not Significantly Different Based on the Honestly Significant Difference Test (BNJ) level 0.05

[2] stated that the character of a plant that is taller and does not touch the ground can reduce water splashing from the soil to the fruit which is one source of disease infection. [10] stated that the fruit from higher plants does not touch the ground so that it can reduce water splashing from the soil to the fruit which is one source of fungal infection.

The P60 treatment showed no significant effect on the variables of plant height and dichotomous height of Aceh's local chili plants. Although the average value of plant height in Aceh local varieties given P60 was (54.61 cm) higher than without P60 (52.40 cm). The average dichotomous height value of chili plants given P60 (24.38 cm) was higher than without P60 (22.96 cm).

3.3. Stem diameter and heading width
The average value of stem diameter and crown width on Aceh local varieties of chili made after the harvest 2 can be seen in Table 3.
Table 3. Average value stem diameter and heading width in Aceh local varieties due to P60 treatment.

| Genotype          | Vegetative Growth |   |
|-------------------|-------------------|---|
|                   | Stem Diameter (mm) | Heading Width (cm) |
| Odeng             | 6.45 a            | 27.87 a |
| Lanyoe            | 8.17 ab           | 37.71 ab |
| S. Lamando        | 8.61 ab           | 37.80 ab |
| LamandoLapaben    | 9.17 b            | 33.80 ab |
| Baja F1           | 8.95 b            | 38.98 b |
| BNJ 0.05          |                   |   |
|                   | 2.34              | 10.46 |

P60 treatment

| P1    | 8.47 | 34.04 |
| P0    | 8.06 | 36.42 |

BNJ 0.05

Note: Numbers Followed by the Same Letter in the Same Column Are Not Significantly Different Based on the Honestly Significant Difference Test (BNJ) level 0.05

The average value of the stem diameter of the local Aceh varieties tested ranged from 6.45 to 9.17 mm. The average width of the canopy of local Aceh chili plants ranges from 27.87 to 38.98 cm. The LamandoLapaben variety had the thickest stem diameter (9.17 mm) which was not statistically significantly different from the comparison variety Baja F1 (8.95 mm). The widest canopy width parameter was found in Super Lamando variety (37.80 cm) which was statistically significantly different from the comparison variety Baja F1 (38.98 cm). The LamandoLapaben variety produces a large average stem diameter, this indicates that the LamandoLapaben variety tends to be able to grow well in a state of begomovirus infection compared to other varieties.

According to [16] resistance to a disease in various plant varieties will not be the same. Resistance to a disease is controlled by resistance genes that are expressed in plant morphology which will support the mechanism of resistance to the disease. [17] stated that one of the reasons why the resistance gene did not appear was that the resistance gene was controlled by several minor genes which were quantitative in nature, which meant that they were influenced by the environment. In this case it is possible that there is a correlation between the variables of plant height, stem diameter and canopy width of chili plants. This is in accordance with the research of [2] which states that there is an appropriate correlation between plant height growth and stem diameter, the higher the plant the higher the dichotomus, the greater the stem circumference and the wider the canopy of chili plants.

P60 treatment showed no significant effect on the variable diameter and crown width Aceh local pepper plant. Although the average diameter of the local varieties of Aceh provided P60 (8.47 mm) than those without P60 (8.06 mm). The average value of chili plant title width given P60 (34.04 cm) was lower than without P60 (36.42).

3.4. Peroxidase enzyme analysis

Peroxidase is an enzyme that can indicate whether a plant is induced by systemic resistance or not by measuring its activity. Table 4 shows that the average value of peroxidase enzyme activity in chilies given P60 ranged from (1.18 – 1.61 Umg-1 protein) while without P60 the value of peroxidase enzyme activity ranged (0.78 - 0.99 Umg-1 protein). The average value of the highest peroxidase enzyme activity was found in the Lamando Lapaben variety with the addition of P60 which the peroxidase enzyme activity value was 1.41 Umg-1 protein compared to the Baja F1 variety, namely 1.61 Umg-1 protein. [18] stated that disease-resistant plants tend to show higher peroxidase activity than susceptible plants.
Table 4. The average value of peroxidase enzyme analysis.

| Genotype               | Peroxidase Enzyme Analysis (Umg-1 protein) |
|------------------------|--------------------------------------------|
|                        | P1    | P0    |
| Odeng                  | 1,18  | 0,92  |
| Lanyoe                 | 1,24  | 0,78  |
| Super Lamando          | 1,25  | 0,81  |
| LamandoLapaben         | 1,41  | 0,87  |
| Baja F1                | 1,61  | 0,99  |

4. Conclusions
The use of P60 has a very significant effect on the incidence of disease and disease incubation. The lowest incidence of disease occurred in the Baja F1 variety (51.95%) and the highest was in the Odeng variety 61.95 (%). The variety treatment had a significant effect on the incidence of disease incubation and vegetative parameters.

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