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Resistant improvement of *Fusarium oxysporum* and production of Aceh local chilli inoculated with rhizobacteria

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**Abstract.** This study aims to obtain information about the induction of local Aceh red chili genotype resistance by using rizobacteria isolates against the disease caused by *F. oxysporum*. This research was conducted from March to September 2018. The design used was a completely randomized design with factorial pattern. The experiment consisted of two factors, the first was red chili genotype, namely the genotype Odeng, Lamando Lapaben, Super Amando 002 and "Lanyoe" SS, and the second factor was rhizobacterial isolate, namely HWI 5 (4) and HWI 8 (6) with a comparison of control treatment. The results showed that the Lanyoe genotype had the highest dichotomous height of 24.44 cm. The combination of the best treatment on the disease incidence parameters, and the number of fruits per plant was found in the combination of the Super Lamando genotype with HWI rizobacteria 8 (6) with each value of 1.00%, and 24.7. The best fruit weights per plant was found in the combination of the Lamando Lapaben genotype with HWI rizobacteria 5 (4) the average value of fruit weight per plant was 12.33 g.

1. Introduction

Chili is one of the vegetables that is needed by the most communities. Chili contains alkaloid compounds such as capsaicin, flavonoids, and essential oils as well as many nutrients such as protein, fat, carbohydrates, calcium (Ca), phosphorus (P), iron (Fe), vitamins A and C which are useful for human health [1].

The increased of chili production is not unstable. Generally, in the rainy season chili production decreases dramatically. The decrease in chili production led to an increase in prices. Chili production decreases during the rainy season due to the high attack of pathogens and has not been obtained by chili plants that are resistant to disease and high production.

Aceh has several local genotypes which has quite high potential is quite high including: Odeng, Lanyoe, Super Amando, and Lamando Lapaben [2]. However, these four genotypes often experience a decrease in production due to pests and diseases. The results showed that the four Acehnese local genotypes were not resistant to disease [3].

*Fusarium oxysporum* is a pathogen that causes wilting in red chili plants. This pathogen can attack red chili plants from the germination period to adulthood. The presence of *F. oxysporum* attacks is one of the limiting factors that causes a decrease in chili production. Losses due to fusarium wilt in chili plants is quite large. [4] States that this disease can cause losses and crop failure up to 50%.
Plant Pest Control carried out by farmers generally still uses synthetic pesticides in the form of fungicides, because farmers consider this method the easiest and most effective. The results showed that the unwise use of synthetic pesticides caused a lot of harm to humans and agroecosystems. For example, synthetic fungicides that pollute the environment have caused human death in the world to reach 40% [5].

The use of biological agents to suppress *F. oxysporum* attacks is certainly a highly recommended choice. One of the biological agents that can be used is by utilizing rizobacteria. Rizobacteria are also capable of triggering plant-induced systemic endurance, thus providing protection against plants from phytopathogen attack. This rhizobacteria ability of is needed to be utilized to prevent and reduce damage caused by plant pathogens [6].

[7] Stated that the results of antagonistic testing with multiple in vitro culture test methods, obtained a number of rhizobacteria as candidates for biocontrol agents that have the ability to inhibit the growth of pathogenic colonies varying depend on the type of isolate type. One of the efforts on improving plant resistance to disease is through the systemic resistance induction triggered by the application of plant growth promoting rhizobacteria (PGPR) as a biocontrol agent [8].

[9] Stated showed that rhizobacteria which inhibit pathogens in vitro were found in isolates HWI 4 (4), HWI 5 (1), HWI 5 (4), HWI 8 (6) and BS3 4 (5) with each each percentage were 52.59%, 54.21%, 56.61%, 57.63%, and 54.01%. As a continuation of previous studies, this study aims to evaluate the effectiveness of HWI 5 (4) and HWI 8 (6) isolates in controlling *F. oxysporum* fungi in vivo. Rizobacteria which are effective in controlling *F. oxysporum* fungus in vivo are used to coat chili seeds (seed treatment) with the aim to control *F. oxysporum* and to increase the growth of chilli plants.

This study aims to obtain information about the induction of local Aceh red chili genotype resistance using rhizobacterial isolates against *F. oxysporum* fungi.

2. Research Method

This research was conducted in March to September 2018. Propagation activities and maintenance of *F. oxysporum* pathogenic fungi were carried out at the Seed Science and Technology Laboratory of the Agrotechnology Study Program at Syiah Kuala University, Banda Aceh.

The planting material used was 4 genotypes, namely the Odeng, Lamando Lapaben, Super Amando 002, and SS "Lanyoe" genotypes, each from Bener Meriah Regency. Other ingredients were pure isolates of *F. oxysporum* and Rizobacteria isolates obtained from the collection of Seed Science and Technology Laboratory of the Faculty of Agriculture, Syiah Kuala University.

The design used is a completely randomized design of factorial patterns. The experiment consisted of two factors, namely the red chili genotype used, namely the genotype Odeng, Lamando Lapaben, Super Amando 002, and "Lanyoe" SS. The second factor was rhizobacterial isolate, namely HWI 5 (4) and HWI 8 (6) with a comparison of control treatment. So that there are 12 treatment combinations with 3 replications consisting of 36 experimental units with 10 plants in each treatment so that a total of 360 experimental units is obtained.

Data analysis using ANOVA. If the F test results show a significant effect of treatment (α = 5%) then the test will be performed between the average of the treatments using a follow-up procedure with a DMRT test of α = 0.05.

Rizobacteria used were isolates from the results of isolation used by previous researchers and were a collection of Seed Science and Technology Laboratory of the Faculty of Agriculture, Syiah Kuala University. The seeds that have been cleaned are then soaked with 150 ml rizobacteria suspenze which has been prepared 1 x 24 hours with a population density of 109 cfu / ml and ready to be used for breeding [9]. Seeds that have received rizobacteria treatment are germinated using a planting medium in the form of a mixture of roasted husk and sterile compost (2: 1) which was previously sieved with 9 mesh sieve. 10 seeds were planted in each treatment in a tray measuring 27 x 56 x 5 cm (length x width x height).

*F. oxysporum* inoculation is carried out when the chili plant is 33 HSS (Days after the seedlings or when the chili plant has two fully exposed leaves. Inoculation is done by soaking the roots of the
seedlings that have been mechanically injured into a 1-hour sandalwood suspension inoculation. Inoculation is carried out in two stages namely, when transplanting is carried out by soaking the roots of seeds that have been injured in the fungus substrate and when the chilli plants are 21 HSP (Days After Transplanting) by means of 10 g of inoculum soil spread around the plant and the base stem injured [10].

The planting medium used is a mixture of soil and manure with a ratio of 2:1. Growing media containing the seeds are placed regularly within 30 cm x 50 cm. Fertilization is done after the plant is 1 week after planting (MST) with fertilizer application using NPK fertilizer solution (16:16:16) at a dose of 10 g L−1, each plant is given as much as 250 ml of fertilizer solution. When applying fertilizer once a week [11]. Variables observed were disease incidence; Dichotomous height; Fruit weight per plant; Number of Planted Fruits

Disease incidence were observed every day along 7 days started one day after inoculation. Observations were made by counting the number of infected plants marked by symptoms of sprouting [12]. To find out the incidence of disease at the observation site calculated using the formula for the incidence of disease

\[ DI = \frac{n}{N} \times 100\% \]

Where:
- DI = disease incidence
- n = Number of infected plants
- N = Number of plants observed [13].

3. Results and Discussion

3.1. Dichotomous Height

In the dichotomous height parameter, the Lanyoe genotype produced the highest dichotomous height of 24.44 cm which was significantly different from the other genotypes. While the super lamando genotype has the lowest dichotomous height of 16.74 cm (Table 1)

Genotypes with high dichotomous that are good will avoid the occurrence of infectious diseases caused by soil borne diseases due to splashes of water on the stems or fruit of the plant. Significant differences in the Aceh local chili dichotomus are expected to result in high genetic diversity coefficients. The higher the genetic diversity coefficients value, the more beneficial it is in the process of crop selection because the characters between genotypes tested are increasingly different and vice versa [14].

Table 1. Average height of the dichotomous four genotypes of local Aceh chili

<table>
<thead>
<tr>
<th>Treatment</th>
<th>height of the dichotomous (cm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Odeng</td>
<td>20.12 a</td>
</tr>
<tr>
<td>Lamando Lapaben</td>
<td>19.83 a</td>
</tr>
<tr>
<td>Super Lamando</td>
<td>16.74 a</td>
</tr>
<tr>
<td>Lanyoe</td>
<td>24.44 b</td>
</tr>
</tbody>
</table>

Note: Numbers followed by the same letter in the same column are different not evident in the 0.05 DMRT Test

The value of genetic diversity coefficient can provide information about the level of genetic diversity of members of the plant population in a plant population that is tested [15].

3.2. Percentage of Disease Incidence

The results showed that administration of rhizobacterial isolates of HWI 8 (6) could induce optimal resilience as indicated by the low percentage of fusarium wilt disease events that occurred. This is in accordance with [16] which states that in vitro rhizobacterial testing of HWI 8 (6) has a high inhibitory value of the pathogenic \( F. oxysporum \) with a percentage of inhibition of 72.22%.
Table 2. Average interactions of genotypes and rhizobacteria on the percentage of disease incidence

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Disease Incidence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No rhizobacteria</td>
</tr>
<tr>
<td>Odeng</td>
<td>3.13 Ba</td>
</tr>
<tr>
<td>Lamando Lapaben</td>
<td>2.85 Aa</td>
</tr>
<tr>
<td>Super Lamando</td>
<td>3.13 Ba</td>
</tr>
<tr>
<td>Lanyoe</td>
<td>2.91 Ba</td>
</tr>
</tbody>
</table>

Note: Numbers followed by the same letter in the same column are different not evident in the 0.05 DMRT Test

[8] Also added that indicators of induction of plant resistance to pathogenic infections caused by rhizobacteria occur due to an increase in the activity of peroxidase enzymes, phenylalanine amonialase and polyphenol oxidase and production of phytoalexin compounds.

The research conducted by [17] also shows the same thing. In this case it was found that the treatment of chilli seeds with rhizobacteria before planting (by pathogen inoculation) can show the same growth with positive control (seed without treatment and without pathogen inoculation). [7] also added that Induced Systemic Resistance (ISR) is one of the mechanisms that occur in rhizobacteria with the ability of antagonism.

3.3. Weight of Fruit each Plant

The results of measurements of fruit weights per plant showed that each treatment combination had a different fruit weight value. This variation is thought to be caused by differences in the treatment of rhizobacteria used.

Table 3. Average fruit weight each plant

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Without Rhizobakteri</th>
<th>HWI 5(4)</th>
<th>HWI 8(6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Odeng</td>
<td>12.28 Ba</td>
<td>9.77 Aa</td>
<td>6.62 Aa</td>
</tr>
<tr>
<td>Lamando Lapaben</td>
<td>10.99 Aa</td>
<td>12.33 Bb</td>
<td>7.21 Aa</td>
</tr>
<tr>
<td>Super Lamando</td>
<td>8.33 Aa</td>
<td>6.56 Aa</td>
<td>10.75 Bb</td>
</tr>
<tr>
<td>Lanyoe</td>
<td>11.18 Aa</td>
<td>8.53 Aa</td>
<td>11.45 Ac</td>
</tr>
</tbody>
</table>

Note: The numbers followed by the same letters (capital letters seen according to rows and small letters viewed according to columns) are not significantly different in the DMRT Test of 0.05 HWI = Hakim Wih Ilang

As a biocontrol agent, rizobacteria have different abilities in reducing *F. oxysporum* infection at the time of fruit formation. The mechanism of systemic acquired resistance (SAR) in induction of plant resistance is characterized by the accumulation of salicylic acid and peroxidase [18]. [19] added that in adult plants *F. oxysporum* infection tends to keep making these plants survive and form fruit but the results are very few and small in size.

3.4. Number of Fruits each Plant

In the parameter number of fruits each plant (Table 4) shows that the average number of fruits per plant is found in the combination of Super Lamando genotype treatment with rhizobacteria HWI 8 (6) with an average number of fruits per plant that is 49.40 which is not significantly different from the combination of Lanyoe genotype treatment with HWI 8 rizobacteria (6), significantly different from the combination of Lamando Lapaben genotype treatment with rhizobacteria HWI 5 (4) and the combination of the Lamando Lapaben genotype treatment with HWI 8 rizobacteria (6), as well as significantly different from other treatment combinations.
States that increasing genetic diversity of chili resistance to disease is studied in quantitative characters including also added that polygenic characters are influenced by several traits within the plant itself: height, age of flowering, age of harvest, weight of planted fruit and number of planted fruit.

The results of observations of the number of fruits per plant in each genotype turned out to be different due to differences in rhizobacterial isolates given. Significant differences are found in the Lamando Lapaben and Super Lamando genotypes. In the treatment without rizobacteria, the average number of fruits was the least found in the super lamando genotype. On the contrary, in the treatment of rhizobacteria giving HWI 5 (4) the super lamando genotype produced the average value of the highest number of fruits compared to other genotypes.

Table 4. Average number of fruits each plant

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Benchmark Results</th>
<th>Without rhizobacteria</th>
<th>HWI 5(4)</th>
<th>HWI 8(6)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Odeng</td>
<td></td>
<td>16.50 Aa</td>
<td>27.83 Aa</td>
<td>29.56 Aa</td>
</tr>
<tr>
<td>Lamando Lapaben</td>
<td></td>
<td>15.61 Aa</td>
<td>49.33 Cb</td>
<td>29.33 Ba</td>
</tr>
<tr>
<td>Super Lamando</td>
<td></td>
<td>12.89 Aa</td>
<td>18.67 Aa</td>
<td>49.40 Bb</td>
</tr>
<tr>
<td>Lanyoe</td>
<td></td>
<td>16.83 Aa</td>
<td>19.83 Aa</td>
<td>43.61 Bb</td>
</tr>
</tbody>
</table>

Note: The numbers followed by the same letters (capital letters seen according to rows and small letters viewed according to columns) are not significantly different in the DMRT Test of 0.05 HWI = Hakim Wih Ilang

Likewise, the comparison of the average value of the number of fruit plants in the Super Lamando genotype without rhizobacteria treatment and with rhizobacteria treatment HWI 8 (6). In this case, it shows that both rhizobacteria provide good resistance response to chili plants under *F. oxysporum*. This is reinforced by the statement of [22] stating that several studies have proven the use of rizobacteria as biocontrol agents can induce increased plant growth and ultimately increase yields as a result of long-term disease control.

4. Conclusion
The Lanyoe genotype dichotomous height showed the highest dichotomous value of 24.44 cm. The best treatment combination on the disease incidence parameters, and the number of fruits per plant was found in the combination of the Super Lamando genotype treatment with HWI rizobacteria 8 (6) with each value is 1.00%, 24.7 and 49.40. The best plant fruit weights were found in the combination of the Lamando Lapaben genotype treatment with HWI rizobacteria 5 (4) with an average value of fruit weight per plant that was 12.33 g. The Super lamando genotype given HWI 8 rhizobacteria has the best resistance to disease and has the highest number of fruit per plant. While the Lamando Lapaben genotype that was given rhizobacteria HWI 5 produced the highest fruit weight per plant. So, for farmers in areas that experience many incidences of disease. It is recommended to use the Super Lamando genotype in combination with HWI 8 Rhizobacteria.

Acknowledgements
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