**ABSTRACT**

Maize production in Madura Island is low. The study aimed to evaluate the agronomic performances (flowering age, harvesting age, and yield), heterosis effect, and resistance to downy mildew disease of F₁ Madura maize hybrids. Ten F₁ maize hybrids and seven respective parents were evaluated at the experimental center of the Agro-Technology Study Program of Agriculture Faculty, the University of Trunojoyo Madura, from July to December 2017. The experiments were arranged in a randomized block design, three replications, with a 50–100 plant population per unit. The parents were T₁, T₁₀, G₁₀, G₁, E₁₀, Td₁₀, and L₁ genotypes having resistance to downy mildew (Peronosclerospora maydis). The results showed that the flowering age of the F₁ Madura maize hybrids ranged 35–39 days, the harvesting age was 74–81 days, and the yield ranged from 2.90–6.40 t ha⁻¹. Three hybrids showed the highest yield, i.e., T₁₁x L₁ = 6.40 t ha⁻¹, T₁₀x L₁ = 5.42 t ha⁻¹, and E₁₀x L₁ = 5.90 t ha⁻¹, and resistance to downy mildew, i.e., T₁₁x L₁ (26.67%), T₁₀x L₁ (26.67%), and E₁₀x L₁ (26.67%). Two hybrids showed the highest heterosis values for yield, i.e., T₁₁x L₁ (65.80%) and E₁₀x L₁ (54.65%). The study suggests that three F₁ Madura maize hybrids (T₁₁x L₁, T₁₀x L₁, and E₁₀x L₁) are prospective to be developed further for high yield and resistance to downy mildew.

[Keywords: agronomic character, heterosis value, Peronosclerospora maydis]

**INTRODUCTION**

Maize is one of the important crops widely cultivated by farmers in Madura, East Java, Indonesia. The maize planting area in Madura is around 360,000 hectares (Amzeri 2018). But, maize yield in Madura is the lowest compared to other maize production centers in East Java. Maize yield at the farm level in Madura in 2016 was only 2 t ha⁻¹, while maize yield in East Java achieved 5.08 t ha⁻¹ (Pusat Data dan Informasi Pertanian 2016; Amzeri 2018).

The causes of the low yield of maize in Madura are: (1) less fertile land, (2) low rainfall, and (3) use of nonsuperior seeds (Amzeri 2009). The ways to solve these problems could be through (1) improving the environmental conditions in which the plant grows and develops, and (2) assembling a variety that is resistant to biotic or biotic environmental stresses and has a high yield potential through breeding programs. The second method is more popular than the first one.

The improved appearance of plants can be through the utilization of heterosis effect. The heterosis phenomenon is the action and interaction of good dominant genes collected in one F₁ genotype as a result of crossing two parents (Amzeri 2015). The use of heterosis in some food crops through the formation of hybrid varieties shows favorable results in cross-pollinating plants (Bairagi et al.)
Heterosis is widely used to improve the adaptability of hybrid varieties. According to Patel et al. (2010), maize yield increased after breeders assembled more hybrid varieties compared to composite ones.

Downy mildew is a major disease in maize that is caused by *Peronosclerospora maydis*. The disease has caused a huge loss to maize farmers. According to Soenartiningsih (2015), Yasin and Zubachtiroddin (2006), yield loss due to downy mildew can reach 100%. One step to control the disease is by using resistant varieties, because it is more stable, economical, and does not cause effects such as poisoning and environmental pollution. Based on the description above, identification of heterosis and disease resistance in maize genotype is needed as a basis for assembling hybrid maize varieties suitable for Madura environmental conditions. The purpose of this study was to evaluate the agronomic performances (flowering age, harvesting age, and yield), heterosis effect, and resistance to downy mildew disease of F₁ Madura maize hybrids.

**MATERIALS AND METHODS**

**Plant Materials**

The study was conducted at the experimental center of the Agro-Technology Study Program of Agriculture Faculty, the University of Trunojoyo Madura, from July to December 2017. The study used ten combinations of Madura maize crossing, i.e. T₁₂ x L₁, T₁₆ x L₁, G₁₀ x L₁, G₁₄ x L₁, E₁₀₂ x L₁, Td₁₄₁ x L₁, T₁₂ x G₁₀, G₁₀ x E₁₀₂, G₁₀ x Td₁₄₁, Td₁₄₁ x E₁₀₂, and 7 parents i.e. T₁₂, T₁₄₁, G₁₀, G₁₄, E₁₀₂, Td₁₄₁, and L₁, so there were 17 genotypes. The characters of the seven parents were shown in Table 1.

### Table 1. Characters of Madura maize parents used in the study.

| Parents   | Origin                 | Characters                                     |
|-----------|------------------------|------------------------------------------------|
| T₁₂       | University of Trunojoyo| Yield = 2.25 t ha⁻¹                             |
| T₁₆       | University of Trunojoyo| Yield = 2.27 t ha⁻¹                             |
| E₁₀₂      | University of Trunojoyo| Yield = 2.12 t ha⁻¹                             |
| E₁₄₁      | University of Trunojoyo| Yield = 2.05 t ha⁻¹                             |
| L₁        | Indonesian Cereals, Maros| Yield = 4.92 t ha⁻¹                           |
| G₁₁₀      | Indonesian Cereals, Maros| Yield = 2.34 t ha⁻¹                           |
| G₁₁₄      | Indonesian Cereals, Maros| Yield = 2.22 t ha⁻¹                           |

Source: Badami and Amzeri (2017); Amzeri and Badami (2019)

**Experimental Design to Measure the Heterosis Value**

The study was arranged in a randomized complete block design, ten combinations of Madura maize crossings and seven respective parents, three replications, so there were 51 experimental units. Each experimental unit was a 10 m x 2 m size, and planting distance was 20 cm x 70 cm. Therefore, the plant population in each unit was 100 plants. Observations were made on 40 randomly selected plant samples from each experimental unit. The observed characters were flowering age, harvesting age, and yield. The yield of 40 samples was then converted to a plant population of one hectare.

### Resistance Assay to Downy Mildew

The experiment was arranged in a randomized complete block design, which consisted of 10 genotypes (10 combinations of the crossing), and three replications, so there were 30 experimental units. The size of the experimental unit was the same as described above, but only 20 plant samples were observed from each unit.

Three weeks before the maize hybrids planted, the downy mildew susceptible maize variety *(G₁₁₁)* was planted in three rows at the edge of and among each block as the spreader plants for the downy mildew diseased source plants. When these spreader plants grew, the plants were inoculated by spraying with the spores of downy mildew following the method of Azrai et al. (2000). When the spreader plants were infected with downy mildew > 50%, then ten hybrid genotypes of maize were planted in the experimental units.

### Data Analysis

Heterosis estimation values were assessed as the middle value of the square of the two parents (mid-parent) by using the formula of Hallauer et al. (2010) and Weber et al. (1970) as following:

\[
\text{Heterosis} = \frac{4 \times \mu_{F_{1}}}{\mu_{P_{1}} + \mu_{P_{2}}} \times 100\% \\
\mu_{F_{1}} = \text{middle value of generation} \\
\mu_{P_{1}} = \text{middle value of both parents}
\]

The disease occurrence (KiP) on the tested maize hybrid plants was observed at 14, 21, 28, 35, and 42 days after planting, following this formula:

\[
\text{KiP} = \frac{A}{B} \times 100\% \\
\text{KiP} = \text{diseased occurrence}, A = \text{number of diseased plants}, B = \text{number of plants observed}
\]
The downy mildew resistance category was calculated following Talanca (2009) as presented in Table 2.

The disease severity (I) on the hybrid plants was assessed at 14, 21, 28, 35, and 42 days after planting following Laksono et al. (2010):

\[ I = \frac{\sum_{i=1}^{n} v_i I_i}{n \times V} \times 100\% \]

where \( I \) = disease severity; \( n \) = number of plants observed with the score to-I; \( v_i \) = plants score to-I; \( N \) = total plant observed; \( V \) = highest crop score.

The plant damaged due to the downy mildew was scored in 0–4 categories (Ginting 2013; Pakki and Mappaganggang 2018), namely 0 = no infection; 1 = light attack, < 10% damage per plant; 2 = medium attack, 10–25% damage per plant; 3 = rather heavy attack, 25–50% damage per plant; 4 = heavy attack, > 50% damage per plant. Disease infection rate was observed at 14, 21, 28, 35, and 42 days after planting and calculated using the formula of Pajrin et al. (2013) as follows:

\[ r = \frac{23}{(r^2 - t^2)} \left( \log \frac{X_2}{1 - X_1} - \log \frac{1}{X_2} \right) \]

where \( r \) = infection rate; 2,3 = constanta; \( X_1 \) = the first inoculum, the first infection to occur; \( X_2 \) = percentage of disease at a certain time; \( t \) = observation time when \( X \) is calculated.

### RESULTS AND DISCUSSION

#### Agronomic Characters

The F₁ Madura maize plants started to flower at 35–39 days, and the harvesting time was at 74–81 days after planting. The maize yield ranged from 2.90 to 6.40 t ha⁻¹ (Table 3). These results indicated that the parent combination significantly affected all the three main agronomic characters observed, i.e. the flowering age, harvesting age, and yield. The shortest flowering age was shown from the \( T_{12} \times G_{10} \), \( G_{10} \times E_{02} \), \( G_{10} \times E_{03} \), and \( G_{10} \times Td_{04} \) crossings. The \( Td_{04} \times E_{04} \) crossing showed the shortest harvesting age, whereas the longest was \( T_{12} \times L_{1} \) crossing.

#### Heterosis Value

Heterosis values for the flowering age of the F₁ Madura maize hybrids greatly varied from -3.90 to 4.48 (Table 3).

| Crossing       | Flowering age (days) | Harvesting age (days) | Yield (t ha⁻¹) |
|----------------|----------------------|-----------------------|----------------|
| \( T_{12} \times L_{1} \) | 38 bc                | 79 b                  | 6.40 c         |
| \( T_{10} \times L_{1} \) | 38 bc                | 79 b                  | 5.42 bc        |
| \( G_{10} \times L_{1} \) | 39 c                 | 81 b                  | 4.99 b         |
| \( G_{14} \times L_{1} \) | 37 bc                | 79 b                  | 5.10 bc        |
| \( E_{02} \times L_{1} \) | 38 bc                | 79 b                  | 5.90 bc        |
| \( Td_{04} \times L_{1} \) | 37 bc                | 79 b                  | 3.50 ab        |
| \( T_{12} \times G_{10} \) | 35 a                 | 75 a                  | 2.80 a         |
| \( G_{10} \times E_{02} \) | 35 a                 | 75 a                  | 3.21 a         |
| \( G_{10} \times Td_{04} \) | 35 a                 | 75 a                  | 2.90 a         |
| \( Td_{04} \times E_{02} \) | 35 a                 | 74 a                  | 3.01 a         |

The numbers followed by the same letter in the same column were not significantly different based on Duncan’s test at \( \alpha = 5\% \).
4). The existence of the positive heterosis value in this study is desirable. The magnitude of the heterosis value in several parent combinations was due to the genetic relationship of parents that relatively far away compared to other parent combinations (Barth et al. 2003; Birchler et al. 2010; Ruswandi et al. 2005).

For the flowering age, the heterosis values of the two hybrids showed negative (Table 4), while for the harvesting age, five hybrids were also negative (Table 5). Madura maize hybrids that had the best heterosis value for harvesting age were those that had a negative value because they had a faster harvesting age than the average of both parents (Bairagi et al. 2002). Five hybrids, i.e. T12 x L1, T16 x L1, G10 x L1, E04 x L1, and Td4 x L1 showed early maturity (Table 5).

The heterosis value of yield of the F1 Madura maize hybrids showed that several cross combinations had heterosis values varying from -7.53 to 65.80% (Table 6). The highest heterosis value (65.80%) was obtained by hybrid resulting from the crossing of T12 x L1, followed by E02 x L1 (54.65%). This value indicated that the two hybrids had a higher yield than the parents. Therefore, both hybrids can be used to assemble varieties that have a high yield.

Heterosis values of the F1 Madura maize hybrids were influenced by genetic diversity and genetic distance of parents used. High heterosis values exceeding the average of its parents indicated broad genetic diversity among individuals in the population (Poehlman 2013; Tulu 2004). The crossing of two parents with a far genetic distance will produce high heterosis, meaning that high heterosis was produced by a crossing between two parents who had many different characters. According to Mangoendidjojo (2007), heterosis was caused by the interaction of several genes, especially dominant genes.

**Resistance to Downy Mildew**

The downy mildew resistance of the F1 Madura maize hybrids varied (Table 7 and 8). The downy mildew disease symptoms were first seen at seven days after planting. However, after the plant became older, i.e., days, the disease did not appear probably in stationary phase (Korlina and Amir 2015; Kim et al. 2017). The experiment showed that ten F1 Madura maize hybrids had a disease occurrence at 42 days after planting ranging from 26.67% to 93.33% (susceptible to very susceptible category).

Based on the data of disease infection rate, the infection rate declined with increasing age of plants probably because the plant tissue becomes older and harder. In older plant tissues, the downy mildew occurs as local symptoms. As a result, the older age of the plants will be more resistant to disease. According to Agrios (2005), the younger the plant was infected, the more quickly the disease develops. The study showed that G10 x Td4 crossing had the highest disease attack.

| Crossing         | F1 | Female parent | Male parent | Heterosis (%) |
|------------------|----|---------------|-------------|---------------|
| T12 x L1         | 38 | 34            | 42          | 0.00          |
| T16 x L1         | 38 | 34            | 42          | 4.00          |
| G10 x L1         | 39 | 33            | 42          | -3.90         |
| E04 x L1         | 38 | 34            | 42          | 0.00          |
| Td4 x L1         | 37 | 34            | 42          | -2.63         |
| T12 x G10        | 35 | 34            | 33          | 4.48          |
| G10 x E02        | 35 | 33            | 34          | 4.48          |
| G10 x Td4        | 35 | 33            | 34          | 4.48          |
| Td4 x E02        | 35 | 34            | 33          | 4.48          |

**Table 4. The heterosis values of flowering age of F1 Madura maize hybrids.**

| Crossing         | F1 | Female parent | Male parent | Heterosis (%) |
|------------------|----|---------------|-------------|---------------|
| T12 x L1         | 79 | 73            | 87          | -1.25         |
| T16 x L1         | 79 | 73            | 87          | -1.25         |
| G10 x L1         | 81 | 73            | 87          | 1.25          |
| G10 x L1         | 79 | 73            | 87          | -1.25         |
| E04 x L1         | 79 | 72            | 87          | -0.63         |
| Td4 x L1         | 79 | 73            | 87          | -1.25         |
| T12 x G10        | 75 | 73            | 73          | 2.74          |
| G10 x E02        | 75 | 73            | 72          | 3.45          |
| G10 x Td4        | 75 | 73            | 73          | 2.74          |
| Td4 x E02        | 74 | 73            | 72          | 2.07          |

**Table 5. The heterosis values of harvesting age of the F1 Madura maize hybrids.**
rate (10.37% per week), while the $E_{02} \times L_1$ crossing had the lowest disease attack rate (3.07% per week) (Table 9).

Various factors attributed to the high disease intensity are (1) poor resistance of the plant to downy mildew, (2) highly virulent pathogen infecting the plant, and (3) environmental conditions at the time of research. According to Soenartiningisih (2015), in resistant plants, pathogen development will be inhibited, and the virulence level will decrease, whereas, in susceptible plants, the process of pathogen development goes well.

In observing the disease severity at 14 to 42 days after planting, three hybrids had the lowest attacking level. The three hybrids were resulted from crossing of $T_{12} \times L_1$, $T_{10} \times L_1$, and $E_{02} \times L_1$. At 42 days after planting, the three hybrids had an attacking intensity by 26.67% (moderately resistant category). According to Hakim and Dahlan (1974) and Pajrin et al. (2013), low disease severity was thought to be because the hybrid had the resistance trait which was regulated by many genes (quantitative character) from one gene chromosome, so that even though climate factors and virulent pathogens support the occurrence of infection, these hybrids still survive from the attack of downy mildew. Furthermore, Aday (1975) and Singburaudom and Renfro (1982) stated that major genes controlled downy mildew resistance so that it was a qualitative trait.

In plant breeding programs, the purpose of crossing among parents is to get hybrids that have superior characters. The characters expected in this research were among parents is to get hybrids that have superior characters. The characters expected in this research were

### Table 7. Average of downy mildew (Peronosclerospora maydis) disease occurrence on $F_1$ Madura maize hybrids at 14-42 days after planting.

| Crossing       | Disease occurrence (%) |
|----------------|------------------------|
| 14             | 21                     | 28                     | 35                     | 42                     |
| $T_{12} \times L_1$ | 10.00                 | 13.33                 | 16.67                  | 23.33                  | 26.67                  |
| $T_{10} \times L_1$ | 6.67                  | 13.33                 | 20.00                  | 23.33                  | 26.67                  |
| $G_{10} \times L_1$ | 26.67                 | 46.67                 | 50.00                  | 56.67                  | 60.00                  |
| $G_{02} \times L_1$ | 30.00                 | 50.00                 | 53.33                  | 56.67                  | 60.00                  |
| $E_{02} \times L_1$ | 13.33                 | 20.00                 | 20.00                  | 23.33                  | 26.67                  |
| $T_{12} \times G_{10}$ | 20.00                 | 26.67                 | 33.33                  | 33.33                  | 43.33                  |
| $G_{10} \times E_{02}$ | 23.33                 | 30.00                 | 40.00                  | 46.67                  | 50.00                  |
| $G_{10} \times T_{12}$ | 43.33                 | 53.33                 | 63.33                  | 66.67                  | 93.33                  |
| $T_{12} \times E_{02}$ | 30.00                 | 40.00                 | 46.67                  | 60.00                  | 66.67                  |
| $G_{10}$ (susceptible control) | 50.00                 | 63.33                 | 66.67                  | 93.33                  | 95.00                  |

### Table 8. Average of downy mildew (Peronosclerospora maydis) severity on $F_1$ Madura maize hybrids at 14-42 days after planting.

| Crossing       | Disease severity (%) | Resistance category |
|----------------|----------------------|---------------------|
| 14             | 21                     | 28                     | 35                     | 42                     |
| $T_{12} \times L_1$ | 3.00                  | 11.50                 | 15.50                  | 19.50                  | 25.50                  | Moderately resistant |
| $T_{10} \times L_1$ | 4.50                  | 11.00                 | 17.00                  | 20.50                  | 24.00                  | Moderately resistant |
| $G_{10} \times L_1$ | 12.50                 | 37.50                 | 41.00                  | 46.00                  | 58.00                  | Susceptible          |
| $G_{10} \times E_{02}$ | 15.00                 | 36.00                 | 42.50                  | 51.00                  | 54.00                  | Susceptible          |
| $E_{02} \times L_1$ | 7.00                  | 12.00                 | 17.00                  | 21.50                  | 24.50                  | Moderately resistant |
| $T_{12} \times G_{10}$ | 12.00                 | 23.50                 | 28.50                  | 31.50                  | 39.50                  | Moderately resistant |
| $G_{10} \times E_{02}$ | 21.00                 | 46.00                 | 51.50                  | 59.00                  | 68.00                  | Very susceptible     |
| $G_{10} \times T_{12}$ | 22.50                 | 43.50                 | 53.00                  | 62.50                  | 89.00                  | Very susceptible     |
| $T_{12} \times E_{02}$ | 19.00                 | 36.00                 | 42.00                  | 51.00                  | 56.00                  | Susceptible          |
| $G_{10}$ (susceptible control) | 22.50                 | 42.00                 | 51.50                  | 59.00                  | 72.00                  | Very susceptible     |

### CONCLUSION

Ten $F_1$ Madura maize hybrids have evaluated for flowering age, harvesting age, and yield, as well as downy mildew resistance. The flowering age ranged 35-39 days, the harvesting age was 74-81 days, and the yield ranged 2.90-6.40 t ha$^{-1}$. Three crosses showed the highest yield ($T_{12} \times L_1 = 6.40$ t ha$^{-1}$, $T_{10} \times L_1 = 5.42$ t ha$^{-1}$, and $E_{02} \times L_1 = 5.90$ t ha$^{-1}$) compared with their parents and resistance to downy mildew ($T_{12} \times L_1 = 26.67\%$, $T_{10} \times L_1 = 26.67\%$, and $E_{02} \times L_1 = 26.67\%$). The highest heterosis values for yield were shown by two crosses, i.e. $T_{12} \times L_1$ (65.80%) and $E_{02} \times L_1$ (54.65%). The study suggests that the three $F_1$ Madura maize hybrids ($T_{12} \times L_1$, $T_{10} \times L_1$, and $E_{02} \times L_1$) can be evaluated further for high yield and resistance to downy mildew.

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