Selection individual on mutant genotype of soybean
\((Glycine\ max\ L.\ merrill)\) in \(M_5\) generation based on resistance of stem rot disease \(Athelia\ rolfsii\) (curzi)

M Rahmah\(^1\), D S Hanafiah\(^{1*}\), L A M Siregar\(^1\) and I Safni\(^1\)

\(^1\)Faculty of Agriculture, Universitas Sumatera Utara, Indonesia
*E-mail: dedek.hanafiah@yahoo.co.id

Abstract. This study was aimed to obtain selected individuals on soybean plant \(Glycine\ max\) L. (Merrill) in \(M_5\) generation based on high production character and tolerance of stem rot disease \(Athelia\ rolfsii\) (Curzi). This research was conducted in Plant Disease Laboratory and experimental field Faculty of Agriculture Universitas Sumatera Utara Medan, Indonesia. This research was conducted from December 2016 to June 2017. The treatments were 15 mutant lines genotypes and Anjasmoro variety. The results showed that some lines mutant genotypes can gave the good agronomic appearance character than Anjasmoro variety on inoculation treatment of stem rot disease. Selection performed on population \(M_5\) produces selected individuals with tolerance of stem rot disease from 100 and 200 Gy population.

1. Introduction

Oktara [1] stated that soybean production in 2015 is known to 963.10 thousand tons, an increase of 8.10 thousand tons compared to the previous year. The increase outside Java Island was 30.41 thousand tons, but in Java decreased 22.31 thousand tons. The increase in soybean production was due to a productivity increase of 0.18 quintal per hectare or 1.16 percent. However, it is known that the harvested area has decreased by 1.80 thousand hectares or about 0.29 percent. The decline in harvested area is also related to global climate change affecting agriculture.

Global climate change affects schedule and cropping pattern in which climate and weather are the determining factors as well as limiting the success of the business to farmers. Climate change is causing several threats in agriculture, one of which is the increase of pests and plant diseases. This will affect the national soybean production.

In Indonesia, soybean \((Glycine\ max\ L.\ Merr.)\) is one of the leading commodities of legumes, however it is susceptible to plant pests and diseases. Some harmful diseases in soybeans mainly due to fungal pathogens include stem rot disease of \(A.\ rolfsii\). Blum and Rodriguez [2] reported that \(A.\ rolfsii\) infections cause low percentage of seeds to germinate, thus reducing plant populations in soybean.

The study of rolfsii from Pudjihartati et al. [3] showed that \(A.\ rolfsii\) (formerly \(Sclerotium\ rolfsii\)) is a soil pathogenic fungus that is necrotropic, and is the cause of collar rot in peanut crops. \(A.\ rolfsii\) in humid environment conditions, also infects branches and leaves near the surface of the soil, and can be a bridge spreading the growth of mycelium to other parts of the plant. \(A.\ rolfsii\) infection in susceptible pea plants in the field can decrease pod yields up to 74%.

The study from Sibarani et al.[4] stated that the productivity of soybean crops can be improved through the use of technology. One of the technological components is the use of gamma ray...
irradiation to obtain new high yielding varieties, and to change the morphological appearance of plants aimed at increasing crop productivity.

The results of this study are expected to reveal various useful information in the development of soybean varieties through low-dose gamma ray irradiation to produce mutant genotypes resistance to stem rot disease of *A. rolfsii* Curzi and high yielding in M5 generation.

2. Materials and methods
This research was conducted at Plant Disease Laboratory and research area of Faculty of Agriculture University of Sumatera Utara, Medan. This research was conducted in December 2016 until July 2017. Soybean seeds were planted mutant M5 genotypes obtained from the selection of M4 generation pedigrees. The control plants were Anjasmoro variety as the control and parent plant. There were 7 groups of genotypes used in plastic housing including M5 (100)-A-25 (180 plants), M5 (100)-A-6 (48 plants), M5 (100)-A-12 (100 plants), M5 (100)-A-17 (65 plants), M5 (100)-A-11 (122 plants), M5 (100)-A-8 (38 plants) and M5 (300)-A-6 (45 plants).

Each genotypes were planting in plastic housing (inoculation with *A. rolfsii* fungus). Data analysis was done by calculating the mean value of each character observed then the mean value of each population is tested by using t test. The parameters were plant height, flowering age, number of primary branches, number of pods per plant, number of seeds per plant, seed weight per plant and disease incidence.

3. Results and Discussion
The results showed that the average of several genotypes with agronomic characters showed different results for each character. The results of M5 (100 Gy) showed that flowering age and plant height characters gave very significant value while the number of primary branches, the number of pods per plant, the age of harvest, the number of seeds per plant, and the weight of seeds per plant did not give significant value. The mean value of agronomic characters on M5 (100 Gy) with Anjasmoro can be seen in Table 1.

**Table 1.** The mean value of agronomic characters on Population M5 (100 Gy) compared to Anjasmoro on inoculated media with *Athelia rolfsii* (Curzi)

| Characters                  | Anjasmoro | M5(100) | t value |
|-----------------------------|-----------|---------|---------|
| Flowering age (days)        | 32.880    | 34.627  | 0.007** |
| Plant height (cm)           | 73.578    | 85.468  | 0.006** |
| Number of primary branches  | 2.560     | 2.902   | 0.185   |
| Number of pods per plant    | 69.760    | 68.045  | 0.773   |
| Harvest age (days)          | 101.080   | 100.586 | 0.099   |
| Number of seeds per plant   | 145.520   | 142.850 | 0.821   |
| Weight seeds per plant (g)  | 23.592    | 24.821  | 0.552   |

Note: * = Significantly different from the Anjasmoro population at the 5% level based on t test
** = Very significantly different from the Anjasmoro population at the level of 1% based on t test

The result of t-test analysis on M5 (200 Gy) compared to Anjasmoro on the inoculated medium of fungus *Athelia rolfsii* (Curzi) showed not significant value to all observed characters. The average value of agronomic characters on M5 (200 Gy) can be seen in Table 2.
Table 2. The mean value of agronomic characters on population M₅ (200 Gy) compared to Anjasmoro on inoculated media.

| Characters                  | Anjasmoro | M₅(200 Gy) | t value |
|-----------------------------|-----------|------------|---------|
| Flowering age (days)        | 32.880    | 33.544     | 0.139   |
| Plant height (cm)           | 73.568    | 77.483     | 0.327   |
| Number of primary branches  | 2.560     | 2.694      | 0.590   |
| Number of pods per plant    | 69.760    | 68.408     | 0.818   |
| Harvest age (days)          | 101.080   | 100.639    | 0.139   |
| Number of seeds per plant   | 145.520   | 142.537    | 0.797   |
| Weight seeds per plant (g)  | 23.592    | 25.841     | 0.292   |

Note: * = Significantly different from the Anjasmoro population at the 5% level based on t test
** = Very significantly different from the Anjasmoro population at the level of 1% based on t test

The result of t test on M₅ (300 Gy) compared to Anjasmoro parent in the inoculated medium of A. rolfsii (Curzi) showed a significant difference in the number of primary branches and the harvest age gave very significantly difference value compared to Anjasmoro. Characters of flowering age, plant height, number of pods per plant, number of seeds per plant, weight of seeds per plant, and weight of 100 seeds showed not significantly difference compared to Anjasmoro. The average value of agronomic characters on M₅ (300 Gy) with Anjasmoro can be seen in Table 3.

Table 3. The mean value of agronomic characters M₅(300 Gy) generation compared to Anjasmoro on Inoculated Media

| Characters                  | Anjasmoro | M₅(300 Gy) | t value |
|-----------------------------|-----------|------------|---------|
| Flowering age (days)        | 32.880    | 34.476     | 0.133   |
| Plant height (cm)           | 73.568    | 71.448     | 0.745   |
| Number of primary branches  | 2.560     | 3.476      | 0.023*  |
| Number of pods per plant    | 69.760    | 82.095     | 0.239   |
| Harvest age (days)          | 101.080   | 102.524    | 0.001** |
| Number of seeds per plant   | 145.520   | 169.286    | 0.328   |
| Weight seeds per plant (g)  | 23.592    | 33.148     | 1.000   |

Note: * = Significantly different from the Anjasmoro population at the 5% level based on t test
** = Very significantly different from the Anjasmoro population at the level of 1% based on t test

The results showed that the appearance of agronomic characters on the inoculated medium of A. rolfsii (Curzi) causes stem rot disease is lower than planting under optimum conditions without inoculation of fungi in population M₅. This is because A. rolfsii is a soilborne pathogen that infect the base of the stem in the soybean crop and forms a white sclerotium that eventually becomes brown. Ferreira and Boley[5] stated that A. rolfsii infects some parts of the plant under appropriate environmental conditions including roots, petioles, leaves and flowers, however it infects stem at the early stage of disease cycle.

Based on the selection of inoculated media of A. rolfsii (Curzi) obtained 62 selected individuals from each genotype with the tolerant character of stem rot disease A. rolfsii (Curzi). The number of selected individuals is more generated from the population of 100 Gy and 200 Gy irradiation. Harsanti and Yulidar[6] stated that the advantages of mutation techniques can alter and improve one character.
of a variety without changing the other character. This change creates a new trait that is not owned by its parent.

The result of observation diagram of incidence of 24 days after inoculation disease can be seen in Figure 1. The results showed that the affected plant from plot 1 were G M100-A-25 (2/7) by 73.1%. G M100-A-25 (3/4) by 77.1%. and G M100-A-25 (3/7) by 76.9%. G M300-A-8 (37/2) by 100%. G M300-A-8 (35/7) by 87.5%. G M300-A-8 (33/8) by 75.0%. G M300-A-8 (33/3) by 100% and Anjasmoro by 80%. The number of affected plant from plot 2 were G M200-A-12 (6/5) by 88.8%. G M200-A-11 (39/7) by 81.4%. G M200-A-11 (32/3) by 67.4% and Anjasmoro by 87.5%. The number of affected plant from plot 3 were G M100-A-6 (30/2) by 84.0%. G M100-A-6 (31/1) by 88.8%. G M200-A-17 (18/5) by 80.6%. G M200-A-17 (13/6) by 76.9% G M100-A-25 (5/3) by 91.7% and Anjasmoro by 100%.

![Diagram of disease incidence at 24 days after inoculation (DAI)](image)

**Note**: DAI: days after inoculation

**Figure 1.** Diagram of disease incidence at 24 days after inoculation (DAI)

The highest percentage of disease incidence was on G M300-A-8 (37/2). G M300-A-8 (33/3) and Anjasmoro (100%). Stem rot disease can decrease the production of soybean crops. The varieties used also depend on the sensitivity or absence of disease infection. This is in accordance with Rahayu[7] who states that Anjasmoro variety in humid environment such as in Genteng-Banyuwangi infected with *A. rolfsii* with a high incidence of disease. which reached 23%. Symptoms of malformation include root rot and the base of the stem, fallen seed (damping-off), wilted, dead plants, and pod rot. Stem rot disease caused by *A. rolfsii* in the field resulted in dying suddenly after entering the generative phase. the initial symptoms at the base of the stem are surrounded by white mycelium and wilted leaves then plants are blackened like burning. The progression of stem rot disease spread very rapidly due to the humid and wet environment conditions in the planting area when the soybean was planted during the rainy season in March. Sclerotium of fungi can spread through water. *A. rolfsii* is one of the soil pathogens in legumes. Xie[8] reported that *A. rolfsii* likes warm conditions. wet weather and continues to spread when hyphae and sclerotia reach the vulnerable plant tissue so that this condition is advantageous to survive.

**4. Conclusions**

The results showed the appearance of agronomic characters on the inoculated medium of *A. rolfsii* more lower than treatment without inoculation of *A. rolfsii* in population M5. Selection
performed on population Ms produces selected individuals with tolerance of stem rot disease from 100 and 200 Gy population.

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