Diverse Morphological Characteristics of Soybean (Glycine max L. Merill) Pods and Seeds Germplasm

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Abstract. Soybean production can be increased through the assembly of new superior varieties. In a plant gene assembly, it is necessary first to discover the characteristics of the parents. One way to find out these characteristics is to conduct a morphological study. The current study employed a randomized complete block design with three replications to 10 soybean lines. The longest young and mature pod was found in MLGG 0582. The thickest young pod was observed in MLGG 0583, while the widest was reported by MLGG 0617. MLGG 0276 had the thickest pod skin of all varieties. In mature plants, the widest and the thickest pods were found in MLGG 0582 and MLGG 0707, respectively. MLGG 0617 reported the longest, the widest, and the thickest seed. There was no positive correlation found among the characteristics of the young pods, except the thickness of the pods and the skin (0.816). However, in the mature plants, the characteristics of the pods were positively correlated with the characteristics of the seeds.

Keywords: Pods morphology, seeds morphology, soybean

1. Introduction

Soybean is a leguminous crop that is widely enjoyed for consumption by the Indonesians in a multitude of forms such as soy sauce, soy milk, tempeh, tofu, and bean sprouts [1]. These food products have become increasingly popular in the western markets because they contain low-calorie protein which is favorable for the vegetarian diet [2]. The high level of plant-based protein contained in these legumes is safe for people’s health [3]. Even, some community members use soybean as a food ingredient for livestock [4]. The vast benefits of soybean have led to an increasing demand for consumable soybean. Unfortunately, soybean production is still considered insufficient to meet social needs.

According to the statistics, Indonesia had to import soybean in 2014 and 2015 since the local production only yielded around 954 and 963 thousand tons of soybean while soybean consumption could amount to 2.67 and 2.77 million tons [5]. Some factors might affect these low yields of soybean, including pest and disease attacks [6], poor seeds quality and growth [7]. There are effective strategies

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that can be applied to overcome these problems. One of which is by assembling superior varieties with high-quality seeds > 2.5 t/ha [8, 9].

Generating superior cultivars is one of the activities of plant breeding that is often done by the plant breeders. The Mahameru variety have proven to be used as an effective parameter to indicate the attack/favorable outcome of soybean inoculation with CPMMV [10]. The MLGG 0751 and MLGG 0753 also were found as useful as a source of genetic variation in assembling an earlier-maturing variety with a high seed yield potential [11].

A collection of genetic resources, also known as a germplasm collection, plays a key role in plant gene assembly. The genetic resources constitute an indicator of the success of assembling a variety [11]. Before the assembly, researchers need to recognize germplasm characteristics [12]. The characterization of the resources can be done by studying the plant’s morphology. The knowledge of plant morphological characteristics can be beneficial in determining the genetic diversity of a plant. A study suggested that there was a difference found in the length, width, ratio, and length of the petiole of a leaf as well as in the plant height in some future CpMMV-resistant varieties and other superior soybean varieties, such as Gumitir and Wilis. The external structure of a plant can help block an attack from pests [13, 14]. The characterization of soybean morphology can be done to the pods and the seeds.

Research on soybean pods and seeds characteristics needs to be conducted due to the nature of pods that can help plant breeders to recognize the high quality parental for plant breeding in the future. Soybean pods and seeds are organs to store assimilates in the form of a seed. It covers the seed and protects it from pests [15]. The morphological characteristics of a pod can be used to determine the preferences of a pod borer and identify soybean lines [16]. The quality of the seeds born in a pod depends on the physical form of the pod. The shape and the size of a pod, as well as the shape and the size of a seed, represent the morphological characteristics of soybean lines [17] and the diversity level of the plant in a certain area [18]. Therefore, the present research aimed to investigate the characteristics of pods and seeds from many soybean lines.

2. Methods

2.1. Research Setting
This research was carried out at the experimental garden of the Nuts and Tuber Crops Research Center in Jambegede, Kepanjen, Malang, East Java, Indonesia (8°05′36.0″S 112°47′54.0″E). The experiment was conducted from April to August 2017.

2.2. Plant Materials
Plant materials used in this study were the seeds of ten (10) soybean lines from Indonesian Legume and Tuber Crops Research Institute, Indonesian Agency for Agricultural Research and Development in this study. They were MLGG 0276, MLGG 0523, MLGG 0582, MLGG 0583, MLGG 0617, MLGG 0707, MLGG 0714, MLGG 0739, MLGG 0745, and MLGG 0757. The plant materials are different in protein content, where the protein content is as follow MLGG 0276 (38.79%), MLGG 0523 (43.66%), MLGG 0582 (45.47%), MLGG 0583 (44.94%), MLGG 0617 (40.63%), MLGG 0707 (40.19%), MLGG 0714 (40.31%), MLGG 0739 (43.94%), MLGG 0745 (43.34%), MLGG 0757 (41.38%).

2.3. Research Design
The experiment employed a randomized group design with three replications. The soybean seeds were grown on a wedded and cultivated land of 2.24 m² with a planting spacing of 40 cm x 15 cm. Each plot was filled with at least two seeds. The plants were regularly fertilized using 100 kg SP36, 250 kg NPK Phonska, and 1 t/ha manure and also intensively controlled to protect them from weed, pests, and diseases.
2.4. Observation Parameters
The morphological characteristics of the young pods, the morphological characteristics of the mature pods, and the morphological characteristics of the seeds were the observed variables of this research. Specifically, the observations were conducted to the length, the width, and the thickness of the pods and the pods’ skin also the length, the width, and the thickness of the seeds. The young pods were observed on day 54 after sowing, and the observation of the mature pods was conducted on day 80 after sowing. A pair of calipers were used to measure the length and width of the pods and the seeds also the thickness of the seeds. Meanwhile, the thickness of the pods and the pods’ skin were measured using a micrometer screw gauge.

The length of the soybean pod (Figure 1) was measured from the tip to the base (a); the width of the pod was measured from left to right (b); the thickness of the pod was determined on the part that contains seeds (c); the length of the seed was measured from the tip to the base (d); the width of the seed was measured from left to right (e); the thickness of the seed was observed from the front to the back (f).

Figure 1. Measuring the length of the pod (a), the width of the pod (b), the thickness of the pod (c), the length of the seed (d), the width of the seed (e), the thickness of the seed (f)

2.5. Data Analysis
The data were analyzed using ANOVA in SPSS 24. If the results reported a significant difference, the data would be further analyzed using BNT and t-test. A correlation test was also performed to investigate the relationships between the observed variables.

3. Results and Discussion

3.1. The Length of the Pods
There was a significant difference found in the length of the young pods and the mature pods among the ten soybean lines. The longest pod in the young plants was observed in MLGG 0582 with an average size of 43.358 mm (Figure 2 left). This soybean lines shared an almost similar size with MLGG 0276 (39.033 mm), MLGG 0707 (40.521 mm), and MLGG 0745 (39.050 mm). The shortest
pod was found in MLGG 0739 (35.596 mm), which differed significantly from that of MLGG 0582 and MLGG 0707. These young pods would continue to grow until they reached 15-20 days of age [19]. The longest mature pod was reported by MLGG 0582 (41.692 mm), and the shortest mature pod was observed in MLGG 0739 (34.325 mm). The size of the pods can be determined by several factors, including pests attack and a lack of nitrogen supply [20-21]. Small size and a wrinkled shape may indicate the condition where a pod has stopped growing which can result in decreasing the quality of the seeds and the production of the soybean.

The results of the t-test indicated that there was no significant difference (0.641) found between the length of the young pods and the length of the mature pods. The young pods (35.5958 mm – 43.3583 mm) were slightly longer than the mature pods (34.3250 mm – 41.6917 mm). According to [15], pods normally grow until 4.358 cm – 4.883 cm. Findings also suggested that there was no variation in the width and the thickness of the pods and the pods’ skin because the pods were still filled with seeds even though the pods had stopped growing. The most dominant color of the young pods was light green. The pods continue to grow gradually. Meanwhile, the mature pods were mostly brown and easily separated from the stem.

![Figure 2](image-url)

**Figure 2.** Variation in the length of the young pods (left) and variation in the length of the mature pods (right). Letters a-c shows the significance of the differences

3.2. *The Width of the Pods*

The width of a pod can be a determinant factor of the pod’s damage. Pods whose width can reach ± 1.23 cm are the most resistant to any kinds of damage [22]. The widest young pod sized 10.347 mm (MLGG 0583) and the narrowest young pod sized 8.154 mm (MLGG 0523) (Figure 3 left). MLGG 0523 bore a resemblance to MLGG 0739 (8.921 mm). In the mature plants, the widest pod was observed in MLGG 0582 (9.441 mm), and the narrowest pod was found in MLGG 0523 (8.075 mm) (Figure 3 right). The results of the t-test also explained the significant difference between the width of the young pods and the width of the mature pods (a significance level of 0.004). The average width of the young pods was 9.534 mm while the average width of the mature pods was 8.954 mm. The biophysical factors of the width and the length of a pod may affect the preference of a pod borer to lay eggs on a soybean plant [23].
Figure 3. Variation in the width of the young pods (left) and variation in the width of the mature pods (right). Letters a-d show the significance of the differences.

3.3. The Thickness of the Pods
The thickest pod (3.642 mm) was observed in MLGG 0617, followed by MLGG 0707 (3.328 mm), MLGG 0745 (3.052 mm), MLGG 0276 (2.992 mm), and MLGG 0739 (2.641 mm). Compared to the other six young lines, MLGG 0582 reported the thinnest pod with a size of 1.743 mm. In the mature plants, the thickest pod was found in MLGG 0707 (5.609 mm), and the thinnest was observed in MLG 0523 (4.746 mm). The thickness of a pod functions to prevent the soybean plant from pest attack [15]. It is difficult for pests to penetrate through a thick pod so that the seeds in it can still be protected.

Figure 4 shows the results of the t-test indicated a significant difference in the thickness of a pod between the young and the mature plants (a significance level of 0.000). The average thickness of a young pod reached 2.614 mm while a mature pod could grow into 5.174 mm on average because it was already filled with the seeds.

Figure 4. Variation in the thickness of the young pods (left) and variation in the thickness of the mature pods (right). Letters a-e show the significant differences.
3.4. The Thickness of the Pod Skins

The thickest skin was found in MLGG 0276 (0.5874 mm) (Figure 5). This size did not differ significantly from those of MLGG 0523, MLGG 0582, and MLGG 0583. The old pods did not report any significant difference in the thickness of the skin which ranged between 0.665 mm – 0.743 mm (coefficient of variation of variation = 10.054%). The thickness of a pod skin is associated with the resistant of the pod to pests and virus. It also determined the quality and the quantity of the seeds yielded by the plant. Pod borers prefer to lay their eggs on a thin-skinned pod [24].

![Figure 5. Variation in the thickness of the young pods’ skin. Letters a-c show the significant differences.](image)

The results of the t-test with a significance level of 0.000 suggested that there was a significant difference between the thickness of the young and the mature pods’ skin. The mature pods could thicken to 0.704 mm while the young pods could only reach 0.484 mm which made them more prone to pest attack. The pod borers can easily strike pods with thinner skin, and it is more likely that the seeds inside get damaged from the borer’s stylets [16, 25].

3.5. The Length of the Seeds

MLGG 0617 (7.563 mm) reported the longest seed of all lines and thus did not differ significantly from MLGG 0707 (7.454 mm) and MLGG 0714 (7.375 mm) (Figure 6). On the other hand, the shortest seed was observed in MLGG 0523 (6.055 mm) which still fell into a normal category of Indonesian soybean seeds size (5.20 mm – 7.28 mm) [26]. Plants’ seeds may grow in different sizes due to genetic variation and the different levels of sunlight intensity [21, 27].

![Figure 6. Variation in the length of the soybean seeds. Letters a-d show the significance of the differences.](image)
3.6. The Width and the Thickness of the Seeds
MLGG 0617 was a soybean line that reported the widest seed (6.325 mm) and did not differ significantly from other lines, except MLGG 0523, MLGG 0582, MLGG 0583, and MLGG 0757 (Figure 7). Meanwhile, the narrowest seed was observed in MLGG 0523 (5.213 mm). The common size of a soybean seed varies between 4.67 mm – 5.78 mm [26], while the average size of the soybean seeds reported by the ten lines ranged between 4 mm-5.087 mm with a coefficient of variation of 9.720%. Indonesian soybean seeds can grow until 6.36 mm – 7.13 mm [28]. The size of soybean seed is one of the aspects that can predict the quality of the food processed from soybeans, such as tofu and tempeh [29]. Soybeans with a large size of seed are more likable than those with a smaller size [30].

A correlation test was performed to examine the relationships between the observed variables which as a result could depict an increase or a decrease in a variable [31, 32]. A positive correlation was found between the width of the pods and the thickness of the pods’ skin ($r = 0.816^{**}$), which suggested that an increase in the width of a pod was strongly related to a decrease in the thickness of the pod skin (Table 1). In the mature plants, the length of seed was found to be correlated with the width and the thickness of the pod. Besides, a positive correlation was also observed between the thickness of a seed, the thickness of a pod, the length of the seed, and the width of the seed. It indicated that an increase in the thickness of a seed could lead to an increase in the thickness of the pod, the length of the seed, and the width of the seed (Table 2).

Table 1. The Results of the Correlation Test on the Young Pods

|                        | The width of the pods | The thickness of the pods | The thickness of the pod skin |
|------------------------|-----------------------|---------------------------|------------------------------|
| The length of the pods (mm) | 0.503                 | -0.003                    | -0.240                       |
| The width of the pods (mm)   |                       | 0.270                     | -0.170                       |
| The thickness of the pods (mm)|                       |                           | 0.816**                      |

Note: ** A significance level of 0.01 (2-tailed)
Table 2. The Results of the Correlation Test on the Mature Pods and Seeds

|                        | The width of the pods | The thickness of the pods | The thickness of the pod skins | The length of the seeds | The width of the seeds | The thickness of the seeds |
|------------------------|-----------------------|---------------------------|--------------------------------|------------------------|------------------------|---------------------------|
| The length of the pods | 0.568                 | -0.045                    | -0.284                         | 0.015                  | 0.081                  | -0.325                    |
| The width of the pods  | 0.436                 | -0.276                    | 0.642*                         | 0.736*                 | 0.203                  |                            |
| The thickness of the pods | 0.490               |                            | 0.858**                        | 0.743**                | 0.799**                |                            |
| The thickness of the pod skin | 0.341            |                            |                                | 0.073                  |                        | 0.409                     |
| The length of the seeds |                      |                            |                                | 0.927**                |                        | 0.817**                   |
| The width of the seeds  |                      |                            |                                |                        | 0.706*                 |                            |

Notes: **A significance level of 0.01 (2-tailed), *A significance level of 0.05 (2-tailed)

It is stated that the size of a pod has a strong relationship with the size of its seeds [30]. This correlation indicates that an increase in the size of a pod may be affected by a surge in the size of the seeds. The results of the current study suggested some diverse morphological characters of the pods and seeds observed in ten soybean lines, excluding the thickness of the mature pod's skin and the thickness of the seeds. The t-test also indicated that there was no significant difference between the length of the young pods and the mature pods, but the width, the thickness and the skin thickness of the young pods differed significantly from those of the mature pods. The thickness of a soybean pod held a positive correlation with the thickness of the young pod’s skin while some characteristics of a mature pod were found to be correlated with the characteristics of the seed. The findings of this research can be considered in the assembly of a new superior variety of soybean [33, 34].

4. Conclusion

The results showed that the longest young and mature pod was found in MLGG 0582. The widest young pods were reported by MLGG 0617, while the thickest young pod was MLGG 0583. MLGG 0276 had the thickest pod skin of all varieties. In mature plants, the widest and the thickest pods were found in MLGG 0582 and MLGG 0707, respectively. MLGG 0617 reported the longest, the widest, and the thickest seed. There was positively correlated between the characteristics of the pods and the characteristics of the seeds in the mature plants. However, there was no positive correlation found among the characteristics of the young pods, except the thickness of the pods and the skin (0.816).

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