The Diversity of Seed Size and Nutrient Content of Lablab Bean from Three Locations in Indonesia

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Abstract— Lablab bean (Dolichos lablab L.) is one of the legume species that have the potential as an alternative food source for Indonesian people. The purpose of this study was to examine the diversity of seed size and nutritional content of six lablab bean accessions found in the Madura, Probolinggo, and West Nusa Tenggara (WNT), Indonesia. Morphological observations of seeds which included parameters of length, width, thickness, and seed weight, while the nutrient content of seeds was obtained from proximate analysis which included assay of fat, protein, and amylose. Data analysis used in this study was one-way analysis of variance and Pearson Product Moment test. The results of the analysis showed that the variety of accessions had a significant effect on all morphological parameters and nutrient levels studied. Besides, the results of data analysis also inform that some morphological parameters and nutrient content have a significant correlation. The findings of this study also show that lablab seeds contain high protein and amylose, so the use of this plant as alternative food in Indonesia is highly recommended.

Keywords— Lablab bean, nutrient content, morphological diversity.

I. INTRODUCTION

The availability of alternative food sources is an essential condition in various developing countries[1]–[3], including in Indonesia. These food sources should have to meet several criteria, such as cheap, easy to cultivate, rich in energy, and high in protein [4]. The declining national food availability and the increasing population growth position this issue as one of the central issues that must be considered by various parties [5]. Therefore, potential local food exploration needs to be carried out intensively [6], one of which is through research activities.

One group of plants that has great potential as a nutrient-rich food source is Legumes [7], [8]. Legumes or beans are plants belonging to the family Fabaceae (Leguminosae) that are capable of producing seeds in the pod [9]. The ability of various Legumes capable of forming symbiotic with an organism that could fix Nitrogen [10] causes the plant cloud to contain high amounts of protein [11]. The content of starch in the cotyledons of Legumes seeds is also able to provide carbohydrates for humans [12]. In this regard, Indonesia is one country that has many local beans that need to be explored more optimally [13].

One of the local beans found in Indonesia is Lablab bean [14]. Apart from Indonesia, these beans can also be found in India [15], [16], Bangladesh [17], Kenya [18]. This bean, also known as the Dolichos bean, has the Latin name Dolichos lablab L., after previously known as Lablab purpureus L. In some countries, the plants that are believed to come from Africa is also known as Hyacinth bean, Pavita, Chicharas, or Auri [19]. In Indonesia, this plant has a specific name such as in Java known as koroueceng, koropedang or korowedhus; in Madura called kacangkomak; and in Sunda, it is called kacangjerijii [14].

Similar to other countries [20], [21], the lablab bean is still not optimally utilized and cultivated as the primary food source of nutrition for the people of Indonesia [22]. The popularity of the bean is also far behind other legumes, such as peanuts, soybeans, and mung beans. The three legumes are beans which are recommended to cultivate by the Indonesian government policy [23]. Lablab beans are less attractive to the public. If examined further, in addition to the potential as a source of nutrition, lablab bean also contains various bioactive compounds [24]. The existence of these bioactive compounds can potentially act as a natural medicine for various diseases [25]. Some of these diseases include diabetes [26], liver disease [27], to obesity [28].

Based on the background that has been conveyed, it can be seen that lablab bean is an alternative food source that is still marginalized in Indonesia. Efforts aimed at popularizing this plant as a food source for the community need to be carried out sustainably. Studies that examine the benefits of this plant, such as the nutrients contained in the seeds of this plant, also need to
be done. Unfortunately, such information is still rarely found. Various studies of legumes in Indonesia are still often only focused on soybeans [29], [30] and peanut [31], [32]. On the other hand, the research that studies lablab bean is still very little with a limited focus of study. Some of these studies also only involve lablab bean, which is only found on Lombok Island [33], [34]. Therefore, the purpose of this study was to determine the morphology and nutritional content of lablab seeds found in several regions in Indonesia.

II. RESEARCH METHODS

Source of materials

The lab bean seeds used in this study were obtained from plants that grew in several regions in Indonesia. Some of these regions, i.e. in the West Nusa Tenggara region, Probolinggo, and Madura. Two accessions from each region were used as research material in this study. The list of accessions and the codes for each accession are presented in Table 1.

Table 1. List of lablab bean accessions used in this study

| Accessions   | Code |
|--------------|------|
| Madura 1     | Mdr1 |
| Madura 2     | Mdr2 |
| Probolinggo 1| PL1  |
| Probolinggo 2| PL2  |
| West Nusa 1  | WNT1 |
| Tenggara 1   |      |
| West Nusa 2  | WNT2 |
| Tenggara 2   |      |

Morphological measurements

The seeds that have been collected at various sampling locations were taken by the Biology Laboratory of the University of Muhammadiyah Malang. In the laboratory, observing seeds from various accessions that have been collected is done. The data examined in this study are quantitative parameter data on seed morphology. These parameters, including length, width, thickness, and seed weight. Data on length, width and thickness have units of centimetres (cm), while data on seed weights have units of a gram (g). Determination of seed weight using analytical scales. The scales used are “Ohaus Pioneer”. On the other hand, seed thickness is measured using a calliper.

Proximate Analysis

Proximate parameters were analyzed in this study, namely the content of fat, protein, and amylose. The ash to amylose content studied was based on not only the sample's wet weight, but also the dry weight of the sample. The fat content analysis procedure uses the procedures based on the Indonesian National Standard [35], protein content was based on AOAC [36], whereas amylose used the IRRI method [37]. These proximate analyzes were carried out at the Balai Penelitian Tanaman Aneka Kacang dan Umbi, Malang.

Data Analysis

Data obtained in this study were analyzed using one-way analysis of variance (ANOVA). Accession was positioned as an independent variable, while seed length, width, thickness, as well as seed weight and fat, protein, as amylose content were positioned as dependent variables. If the results of the hypothesis test conclude there are significant differences; then the data analysis is continued with the Duncan test with a significance level of 5%. Also, a correlation analysis using Pearson Product Moment was conducted to determine whether there was a relationship between the morphological and nutritional parameters measured in this study.

III. RESULTS AND DISCUSSIONS

In this study, six lablab bean accessions spread across several regions in Indonesia have been collected. The picture of the six lablab bean accessions observed in this study is presented in Figure 1. Furthermore, to determine whether there are differences in seed size and nutrient content of the six accessions, the measurement data are analyzed using one-way ANOVA. The results of the one-way ANOVA tests that have been carried out are presented in Table 2. Based on Table 2, it can be seen that morphological parameters that indicate a significant difference between lablab bean accessions. is the parameter of seed length [F (5,6) = 18.291, p = 0.001], seed weight [F (5,6) = 6.486, p = 0.021], seed thickness [F (5,6) = 5.798, p = 0.027], and seed weight [F (5,6) = 16.736, p = 0.002]. Furthermore, significant differences also occurred in the nutrient content of the seeds, namely the fat content [F (5,6) = 18.485, p = 0.001], protein [F (5,6) = 19.157, p = 0.001], and amylose [F (5,6) = 298.841, p <0.001]. Thus, the ANOVA results showed that the differentiation of seeds from various accessions of lablab bean found in several regions in Indonesia.
Fig. 1: Lablab bean accessions used in this study: a) WNT1, b) WNT2, c) Mdr1; d) Mdr2, e) PL1, and f) PL2

Table 2. ANOVA test results of the effect of accession on the morphological character and nutritional content of lablab seeds

| Dependent Variables | Source | Type III Sum of Squares | df | Mean Square | F     | Sig. | Partial Eta Squared |
|---------------------|--------|-------------------------|----|-------------|-------|------|---------------------|
| Length              | Accession | 0.419                    | 5  | 0.084       | 18.291| 0.001| 0.938               |
|                     | Error    | 0.028                    | 6  | 0.005       |       |      |                     |
| Width               | Accession | 0.189                    | 5  | 0.038       | 6.486 | 0.021| 0.844               |
|                     | Error    | 0.035                    | 6  | 0.006       |       |      |                     |
| Thickness           | Accession | 0.186                    | 5  | 0.037       | 5.798 | 0.027| 0.829               |
|                     | Error    | 0.039                    | 6  | 0.006       |       |      |                     |
| Weight              | Accession | 2.344                    | 5  | 0.469       | 16.736| 0.002| 0.933               |
|                     | Error    | 0.168                    | 6  | 0.028       |       |      |                     |
| Fat                 | Accession | 0.239                    | 5  | 0.048       | 18.485| 0.001| 0.939               |
|                     | Error    | 0.016                    | 6  | 0.003       |       |      |                     |
| Protein             | Accession | 23.543                   | 5  | 4.709       | 19.157| 0.001| 0.941               |
|                     | Error    | 1.475                    | 6  | 0.246       |       |      |                     |
| Amylose             | Accession | 10.049                   | 5  | 2.010       | 298.841| <0.001| 0.996               |
|                     | Error    | 0.040                    | 6  | 0.007       |       |      |                     |

Furthermore, the data were analyzed using the Duncan test. The summary of posthoc test results for morphological parameters is presented in Figure 2. In the Duncan test results graph, accessions with the same alphabet label show no significant difference at the significance level of 5%. Based on Figure 2, it can be seen that the accession of WNT 2 produces the longest seeds. However, the seed length of the accession was not significantly different from the accession of Madura 1 and WNT 2. On the other hand, accession of Probolinggo 2 had a significantly shorter size compared to all accessions collected in this study. Also, it can be seen that Probolinggo 1’s accession has widest seeds. However, the width of these seeds is not significantly different from the accession of Madura 1 and WNT 2. On the other hand, accession of Probolinggo 2 has seeds with the smallest size although the width of the accession is not significantly different from the accession of Madura 2 and WNT 1.
Fig. 2: Duncan test results of the effect of accession on the lablab seeds size

In the parameters of seed thickness, accession of Probolinggo 1 has seeds that are significantly thicker when compared to other accessions, whereas accession of Probolinggo 2 has the thinnest seeds. The accession has seeds whose thickness does not differ significantly from Madura 1’s accession. Furthermore, in seed weight parameters, Probolinggo 1 and WNT 2 accessions have the heaviest seeds, while Probolinggo 2 accessions produce the lightest seeds. The accession has a weight that is not significantly different from the two accessions from Madura.

Fig. 3: Duncan test results of the effect of accession on the nutrition content in lablab seed

Furthermore, the Duncan test results on the parameters of nutrient levels are presented in Figure 3. Based on Figure 3, seeds that have the highest fat content and protein content are accessions of Probolinggo 2 and Madura 1, respectively. The accession of Madura 1 also has the highest amylose content even though Amylose levels were not significantly different from Probolinggo 1 accession. On the other hand, two accessions from WNT and Madura 1 accession had the lowest fat content. In protein parameters, Probolinggo 1 accession has the
lowest levels even though these levels do not differ significantly from Madura 2 and WNT 2 accessions. Finally, Madura 2, Probolinggo 2, and WNT 1 accessions have significantly lower amylase levels than the other three accessions.

Table 3. The correlation test results between the size of the seed and the nutritional content of lablab seeds

|        | Fat   | Protein | Amylose | Length | Width | Thickness | Weight |
|--------|-------|---------|---------|--------|-------|-----------|--------|
| Fat    | 1     | -0.541  | -0.144  | -0.500 | -0.424| -0.333    | -0.441 |
| Protein| -0.541| 1       | 0.179   | 0.114  | 0.093 | -0.440    | -0.212 |
| Amylose| -0.144| 0.179   | 1       | 0.823**| .815**| 0.157     | 0.563  |
| Length | -0.500| 0.114   | 0.823** | 1      | 0.835**| 0.470     | 0.750**|
| Width  | -0.424| 0.093   | 0.815** | .835** | 1     | 0.411     | 0.642**|
| Thickness| -0.333| -0.440  | 0.157   | 0.470  | 0.411 | 1         | 0.737**|

**. Correlation is significant at the 0.01 level (2-tailed).
*. Correlation is significant at the 0.05 level (2-tailed).

The results of the correlation analysis are presented in Table 3. Based on Table 3, it can be seen that some parameters have a significant correlation, while some other factors do not have a significant correlation. The parameters that have a significant correlation, namely between the length of the seeds with amylase content (r = 0.823; p <0.01), the width of the seeds with amylase content (r = 0.815; p <0.01), seed weight with seed length (r = 0.835; p <0.01), seed length with seed weight (r = 0.750; p <0.01), width with seed weight (r = 0.642; p <0.01), and seed thickness with seed weight (r = 0.737; p <0.05).

Overall, the results of this study indicate that differences in accession produce a variety of characters from seeds. The diversity of characters does not only occur in morphological character but also the nutritional content. The emergence of character diversity in the lablab bean species can be caused by genetic factors[38]. As is well known, even in the same species, one member of the species with other members have different genetic variations[39]. The genetic variation refers to variations in allele pairs, thus determining the genotype of each accession[40]. The constitution of the genotype will determine what character will be expressed by the individual[39], [41].

In addition to genetic factors, the environment also plays a role as a factor in the emergence of interspecific diversity[38]. Environmental factors can cause interspecies diversity in at least two ways. The first way is that environmental factors do not cause constitutional genetic changes but will affect the genetic regulation of living things[42]. Certain environmental conditions can suppress the expression of several genes and activate the expression of several other genes[39]. As a result, even though they have the same allele, the characters that appear can be different. The second way, environmental factors will influence the physiological and metabolic processes of living things. As is well known, the environment plays a role in providing nutrients for plants through not only nutrients contained [43], [44] but also microbial activity in the soil[45]. These substances often act as precursors in various metabolisms that occur in cells. Certain environmental conditions were indicated can inhibit the process of nitrogen fixation in the symbiotic activity that occurs in the roots of Legumes[46].

Related to nitrogen fixation, one of the critical nutrients needed by plants is Nitrogen[47]. Nitrogen acts as a primary component of amino acids[48], the monomer of protein. If the Nitrogen level in each location has a significant difference, it is possible that the levels of a protein produced by each plant in these locations are different. Also, environmental factors, such as Nitrogen and carbon dioxide level in the environment affect the photosynthesis rate of plants[49], [50]. Photosynthesis is an anabolic process aimed at producing starch as a food reserve from plants[47]. When the photosynthesis rate is low, the growth and nutrient contained in plants are also low. Given this study involves three regions from three different islands, different environmental factors have been described in this discussion might happen. However, further research needs to be done to ascertain which factors have the most role in the diversity of the phenotype recorded in the results of this study.

Moreover, the proximate test results show that protein is a nutrient with the most content contained in seeds when compared to fat and amylase. These protein levels range from 20.06 to 24.16% from the wet mass of the seed. This range is in line with the previous report that uses lablab bean from local markets in Bangladesh[17]. Therefore, this finding confirms that lablab bean has excellent potential as a protein-rich alternative food
source. Besides have a high protein level, proximate results showed high levels of amylose contained in lablab seeds. The range of amylose levels is from 12.79 to 14.93% of seed’s wet mass. Amylose is the part of the starch, polysaccharides found in seeds that act as an energy source [51]. Amylose forms about 20-30% of the starch structure. Therefore, amylose levels describe how much energy can be obtained from these seeds if consumed by someone. The high amylose levels reported in this study are in line with the high levels of carbohydrates contained by lablab seeds studied in India [16] and Bangladesh [15]. Both studies reported that the carbohydrate content in lablab seeds could reach 48 to 61% of the seed mass. This information proves that the lablabseeds also have the potential to overcome the problem of malnutrition that occurs in some regions of Indonesia.

IV. CONCLUSION
In this study, the diversity of morphology and nutrient levels of six lablab bean accessions was assessed. The results of data analysis concluded that differences in accession caused differences in length, width, weight, and thickness of seeds. Differences in accession also cause differences in levels of fat, protein, and amylose contained in lablab seeds. The results of this study indicate that lablab bean can be used as a functional food because of the high levels of protein and amylose contained in these legume seeds.

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