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Epidermal Structure and Leaf Stomata of Several Accessions of Banana Plants (Musa spp.)

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Abstract: This study aimed to determine the structure of epidermal cells and leaf stomata in Jarum, Merah, Tanduk, Kepok, and Pei or wild banana. The type of research used was quantitative to calculate the number of epidermis, epidermal cell size, number of stomata, and stomata index. Meanwhile, qualitatively described the structure of epidermal cells, epidermal cell shape, stomata shape, and stomata type of banana leaves based on the longitudinal incision method. The results showed that four accessions of bananas have epidermal cell shapes: rectangular, pentagon, hexagon, heptagon, octagonal, and regularly length. Meanwhile, Merah have different epidermal cell shapes, namely rectangular, pentagon, hexagon, and irregular, with untidy arrangements. The stomata of the five samples are surrounded by four to six subsidiary cells and are kidney-shaped. Based on the location of the leaf surface, stomata in five banana accessions have an amphistomatic type. Pei or wild bananas have a long epidermal size compared to the other four species but have a small number of epidermis. Then, the number of stomata in all banana species differs on the upper and lower surfaces. The number of stomata is more on the lower surface of the leaves. Stomata index in all banana species is low.

Keywords: Banana; Epidermal cells; Stomata

Introduction

Indonesia is a country that has a high diversity of flora. Various kinds of plants are found in Indonesia. One of which is banana; there are almost no areas in Indonesia without them (Rai et al., 2018; Saiya et al., 2020). Bananas are one of the leading commodities and make the biggest contribution to national fruit production. In addition to their delicious taste, bananas also contain nutrients, vitamins, and calories, which are beneficial for health (Prahardini et al., 2016).

Generally bananas are known from their fruit parts, but this plant has other parts that are important for growth and development including root, pseudostem and leaves (Zou et al., 2022). Leaves play an important role in maintaining plant survival. Leaves have a stomata structure (leaf mouth), which exchange O_2, CO_2, and water vapor from the leaves to the natural surroundings and vice versa (Sumardi et al., 2010). Stomata function is very important in photosynthesis, respiration, and transpiration. Stomata are found on the upper or lower side of the leaf or only on the lower surface. The number of stomata per mm^2 is different in each plant (Zhao et al., 2006; Hong et al., 2018).

One of the anatomical structures of leaves that receive direct influence from the environment is the epidermal cell. Epidermal cells are the outermost layer of leaves, flowers, fruits, seeds, stems, and roots before secondary thickening. The leaf epidermis of different plants varies greatly in the number of layers, shape, structure, arrangement of stomata, appearance and arrangement of trichomes, and the presence of specialized cells (Casson et al., 2008; Liu et al., 2015).

Among the epidermal cells, there are derivatives, including stomata, trichomes, bulliform cells, silica cells, and cork cells (Liana et al., 2017). Stomata is one of the derivatives of epidemic cells that have important functions for plants. Epidermal cells and stomata are closely related, so environmental factors that affect the epidermal cells will also affect the stomata (Herrmann et

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al., 2021). Leaf epidermis and stomata is one of the important parameters for the field of taxonomy in explaining the systematics of the family level in plants (Ullah et al., 2021). The aimed of this study to determined the structure of epidermal cells and stomata as a potential character of banana accessions.

Method

The plant materials used in this study were five banana accessions (Table 1).

Table 1. List of Five Banana Accessions

Local name/species	Genomic group	Locality
Jarum	AA	Ambon
Merah	AAA, red subgroup	Ambon
Tanduk	AAB, plantain subgroup	Ambon
Kepok	ABB, saba subgroup	Ambon
Pei/*Musa acuminata* var malaccensis	AA	West Seram

The leaf epidermis and stomata were prepared based on the modified Ruzin's method (1999) (Ruzin, 1999). a) The leaves are cleaned with tissue, cut into three parts, and slashed using a sharp blade (the incision obtained must be very thin). Leaf incisions on the lower and upper surfaces of the leaves include incisions at the leaf tip, the leaf center, and the leaf base. b) The incision is immersed in chlorox (Bayclin) for 5 minutes to look white. Then, the incision is removed using a pin and washed in distilled water. c) The leaves are removed and soaked in 1% safranin for 1 minute to color the incisions so that it is easy to see the difference between the stomata and the epidermis. The incision was then washed in distilled water. d) The incision results are placed on a slide, dripped with glycerin, and covered with a cover slip. e) The preparations were observed under a microscope with a magnification of 400x.

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Observational data obtained were then analyzed quantitatively and qualitatively. Mathematically, the formula for the stomatal index is as follows (Siregar et al., 2015), as equation 1.

$$IS = \frac{S}{S+E} \times 100\ \% \quad (1)$$

IS : Stomata index
S : number of stomata in one field of view at 400x magnification
E : the number of epidermal cells in one field of view at 400x magnification

Result and Discussion

Observations of Epidermal and Stomata Cells nn The Upper Surface of The Leaves of Five Banana Accessions

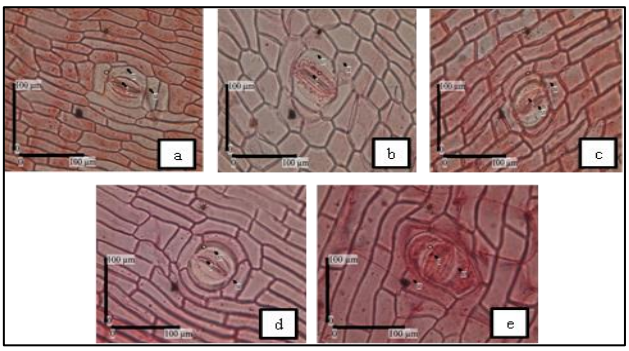

Figure 1. Epidermis and stomata cells on the upper surface of the leaf. (a) jarum, (b) merah, (c) tanduk, (d) kepok, and (e) pei with 400x magnification.

Based on Figure 1, it can be explained that the arrangement of epidermal cells from the Jarum, Tanduk, Kepok and Pei observed were neatly arranged and lined up parallel to the same epidermal cell shape, namely quadrangle, pentagon, hexagon, heptagon, octagon, and a regularly length. Merah have epidemic cell shapes that are different from the four banana accessions observed: rectangular, pentagon, hexagon, heptagon, and irregular shape with the irregular arrangement. The stomata of five banana observed were surrounded by 4-6 epidermal cells with two parallel cells on each side, on the right and left. Epidermal cells surrounding the stomata are called subsidiary cells. The stomata shape of the five samples observed is kidney-shaped.

Observation of Epidermal And Stomata Cells On The Lower Surface of The Leaves of Five Banana Accessions.

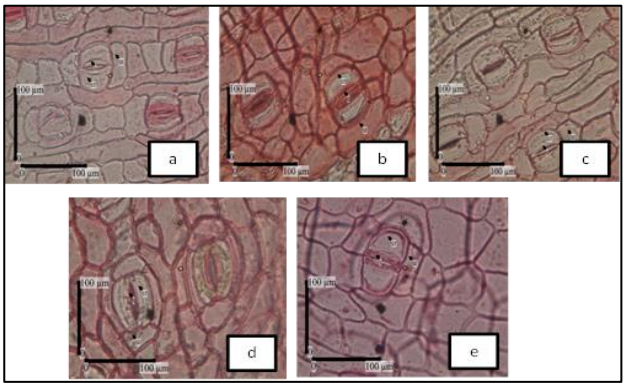

Figure 2. Epidermis and stomata cells on the lower surface of the leaf. (a) jarum, (b) merah, (c) tanduk, (d) kepok and (e) pei with 400x magnification (SE = Epidermal Cell; ST = Neighbor Cell; S= Stomata)

Based on Figure 2, it can be explained that the arrangement of epidermal cells of five bananas observed was the same as on the upper surface of the leaf.

The Average Number of Epidermis, Number of Stomata, And Stomata Index of Five Bananas

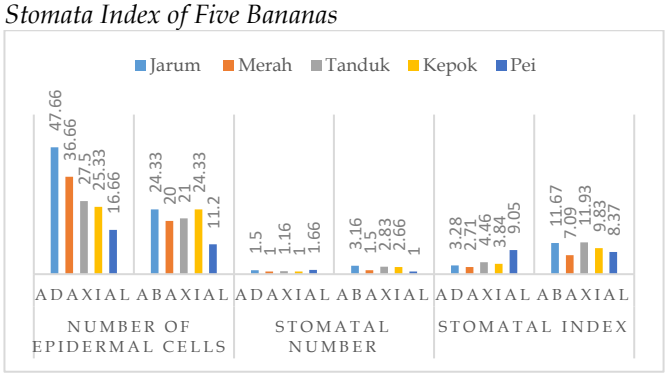

Figure 3. Graph of the average number of epidermis, number and index of stomatal

Based on Figure 3, it can be explained that the Jarum has an average value of a large number of epidermis on the adaxial and abaxial sides, namely 47.66 and 24.33. In contrast, the average value of the small number of epidermis on the adaxial and abaxial side is owned by Pei or wild bananas, namely 16.66 and 11.2. The results of the calculation of the average value of the stomatal index for all bananas observed showed that the abaxial side had an average stomatal index value greater than the adaxial side.

The Average Length and Width of The Epidermal Cells and Stomata of Five Banana Accessions

The study's results on the average length and width of the epidermis and stomata of five banana species showed that the highest average epidermal length on the adaxial and abaxial sides was found in wild bananas with values of 27.05µm and 35.12 µm. In contrast, the lowest average value on the lowest adaxial side of 18.48 µm was found in red bananas. The highest average value of stomata length on the adaxial side was found in red bananas and horn bananas, while the lowest stomata length was found in needle bananas. The average value of stomata length on the adaxial side is relatively the same.

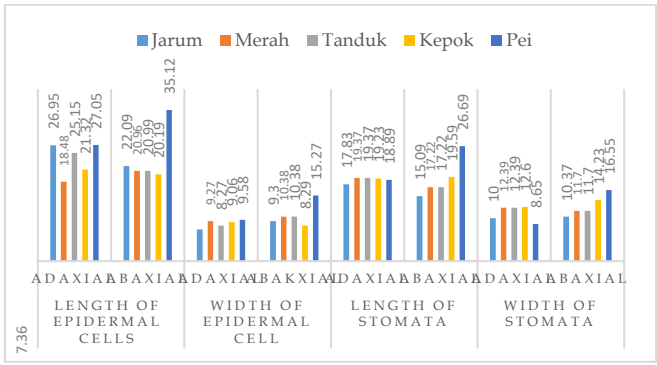

Figure 4. Graph of the average size of epidermal and stomata cells

The study results show that Jarum, Merah, Tanduk, Kepok, and Pei or wild bananas have stomata on the adaxial and abaxial sides. Furthermore, it shows that five banana accessions have an amphistomatic type based on the stomata's location on the leaf surface (Narahayaan et al., 2022; Sumardi et al., 2010). These five samples are monocot plants with the same type of stomata, namely the first type stomata, because the guard cells are surrounded by 4 to 6 subsidiary cells. This condition was in line with Sumardi et al. (2010) that observed Musacea and Rubiaceae family (respectively). This type is usually found in *Araceae, Commelinaceae, Musaceae, Strelitziaceae, Cannaceae,* and *Zingiberaceae* (Rudall et al., 2017; Givnish et al., 2018).

The observations results of the size of epidermal cells greatly affect the number of the epidermis in a wide field of view; if the size of the epidermal cells is small, the number of epidermal cells is more than species that have a larger epidermal size (Asl et al., 2011; Patel et al., 2021). For example, red banana species have a very small epidermal size. However, the number of epidermis is very large compared to wild bananas, which have a large epidermis size but few (Figure 4). The changes in the size of the epidermal cells observed follow the research of Mohapatra et al. (2016), which states that if the light intensity is low, the size of the epidermal cells increases, resulting in a small number of the epidermis.

Based on the results of observations (Figure 3), it was found that there were more stomata on the lower surface of the leaves of Jarum, Merah, Tanduk, Kepok, and Pei or wild bananas. Turner et al. (2007) evaluated

stomata which abundant on the lower surface of the leaf. It is suspected that plants have to maintain their body temperature so that they are not too hot due to direct sunlight, so there are more stomata on the lower surface than on the upper surface. In addition, the upper surface of the leaves is exposed to direct sunlight, so it is not good for plants to have many stomata on the upper surface because it can cause water loss. Therefore, the stomata are on the lower surface of the leaves. According to Ekawati et al. (2014), this may be influenced by plant physiological activities by avoiding stomata from direct exposure to sunlight so that more stomata are found below the leaf surface. The same thing was stated by Li et al. (2017), that the stomata in most plants are more concentrated on the lower surface of the leaves, which reduces transpiration because the lower surface receives less sunlight than the upper surface.

According to Xu et al. (2016), the size of stomata greatly affects the number of stomata. If the size of the stomata is large, the number of stomata is small and vice versa. Growth factors in an environment with high light intensity result in large stomata and a small number of stomata. For example, wild bananas have long stomata sizes but with few stomata. Zhu et al. (2018) reported that the number of stomata can be classified into: few (1-50), quite numerous (51-100), numerous (101-200), very numerous (201->300), and infinity (301->700). The results showed that the number of stomata in all banana cultivars was included in the few criteria (1-50), which ranged from 1-5. It is thought to be due to several factors, such as place, light, temperature, and transpiration. Chatterjee et al. (2020), stated that transpiration activities are affected by external and internal factors. External factors are wind speed, light, water, humidity, and temperature, air pressure. Internal factors are leaf thickness, number of stomata/mm2, presence of cuticle, number of trichomes, and the shape and location of stomata on the surface. Haworth et al. (2015) stated that unique stomatal morphology and structure expressing environmentally sensitive physiological and developmental functions.

Epidermal cells, which are subsidiary cells, do not have chlorophyll, while the stomata guard cells contain chlorophyll, organic phosphate, and phosphorylase enzyme, and in the morning, there is still a little starch in them. The results of measuring the length and width of the stomata of the five banana species showed differences in size. The difference in size is most likely due to genetic factors being more dominant than environmental factors. It is in line with the statement of Meriko (2017) that genetic and environmental factors influence the number and size of stomata. Jarum, Merah, Tanduk, and Kepok are included in the criteria of less long < 20 µm) which is between 17.83-19.37 µm, while wild bananas are included in the criteria for very long (> 25 µm m), namely 26.69 µm. Narahayaan et al. (2022) stated that the length of stomata is less long (< 20 µm), long (20-25 µm), and very long (> 25 µm).

The stomata index shows the ratio between the number of stomata divided by the number of stomata and the epidermis. Compared with many epidermal cells, a small number of stomata will result in a low stomata index and vice versa. If a large number of stomata is compared with a small number of epidermal cells, it will produce a high stomata index (Driesen et al., 2020). The stomata index in all banana species has a low average value of the stomata index (Figure 3). Šantrůček et al. (2014) stated that transpiration activities are affected by external and internal factors. External factors are wind speed, light, water, humidity, temperature, is air pressure. Internal factors are leaf thickness, number of stomata/mm^2, presence of cuticle, number of trichomes, and the shape and location of stomata on the surface. These factors result in a low stomatal index. Sulistiana et al. (2016), the stomata index has several categories, namely the low stomata index (< 20), medium stomata index (20-30), and high stomata index (> 50). The results showed that the average value of the stomata index in all banana species was low because the stomata index ranged from 2.71-11.93.

Conclusion

Based on the results of the research, it can be concluded that 1) The arrangement of the epidermal cells of Jarum, Tanduk, Kepok, and Pei or wild bananas was neatly arranged, namely rectangular, pentagon, hexagon, heptagon, octagonal and regularly length. Merah have a different shape of epidemic cells, which are rectangular, pentagonal, hexagon, and irregular epidemic cells with an untidy arrangement. 2) Wild bananas have a longer but less epidermis than the other four species. 3) Based on the morphology of the stomata type in these samples is type 1. Based on the location of the stomata leaf surface, five banana species have an amphistomatic type. 4) The number of stomata is more found on the leaf's lower surface than on the upper surface. 5) All banana species have low stomata index.

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