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Study of Stomata Characteristics of Plantain and Horn Plants AAB genome

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Abstract: The study of stomata characteristics of plantain and horn plants has been carried out. This study aimed to compare the characteristics of plant stomata by plantain and horn banana AAB genome. Therefore, it is possible to find out the similarities and differences in characteristics between plantain and horn banana that have the same genome, including by stomata location, cover cell shape, stomatal type, number of epidermal cells, number of stomata cells, stomata length, stomata width, stomata area, stomata index, and stomatal density. Stomata observations were made by making an incision using the whole mount method. The results obtained were then analyzed descriptively qualitatively by describing the stomata characteristics of the plantain and horn banana plants.

Keywords: Characteristics; Similarity; Diverensity

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Introduction

Indonesia is one of the world's centers of biodiversity. Biodiversity plays an important role in national development as a biological resource (Basyuni et al., 2018; Verstegen et al., 2019). One of the plants with a high level of genetic diversity is the banana. Indonesia is one of the primary centers of banana diversity. This plant is one of the tropical fruit-producing plants well-known in Indonesian society. This plant also produces fruit that can be consumed, either as fruit eaten directly or as food that must be processed first (Maryani et al., 2019). Types of bananas that are popular in Indonesia include 'Yellow Ambon' with (AAA) Genom, 'Green Ambon' (AAA), 'White Ambon' (AAA), 'Barangan' (AAA), 'Berlin' (AA), 'Lampung' (AA), 'Mas' (AA), 'Raja' (AAB), and 'Raja Sereh' (AAB), while plantain bananas are bananas that are consumed after processing the fruit, namely 'Tanduk' (AAB) bananas, ' Uli' (AAB), 'Kepok' (BBB) and 'Siam' (ABB) (Ningsih & Megia, 2019).

In general, the characteristics possessed by bananas with genome A are small fruit, thin skin, and golden yellow color. As for the banana group with genome B, the characteristics of the fruit are large, thick-skinned, and orange in color. In addition, several types of bananas from genome B have a reasonably high starch content, so they must be processed first before being consumed (Hapsari & Lestari, 2016; Sunaryo et al., 2019).

Banana morphology includes plant parts such as roots, stems, leaves, flowers, and fruit. The growth of these plant parts is interconnected (Padam et al., 2014; Andrew et al., 2021). There are small holes called stomata on the leaves, commonly known as leaf mouths. Stomata are oval surrounded by two guard cells, stomata are also found in almost all parts of the plant body, but the highest number is found in young leaves (Turner et al., 2007; Zait et al., 2017)

The function of stomata is as a place for CO2 to enter from the air in the process of photosynthesis, a place for respiration and transpiration so that stomata are an essential tissue in plant metabolic processes, controlling water loss that is also very important to avoid leaf dehydration due to excessive transpiration (Xu et al., 2016). Stomata are also limited by two special

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epidermal cells, namely guard cells. They consist of a pair of cells that look symmetrical, generally kidney-shaped. On the upper and lower cell walls, there are ledges. The guard cells consist of a pair of cells that look symmetrical, generally kidney-shaped, in the upper and lower cell walls there is also a tool shaped like a ledge. Sometimes the ledge is only found on the upper cell wall.

Neighboring cells are adjacent to or around the guard cells or can also surround the guard cells. The neighboring cells may consist of two or more specifically carry out their functions by associating with guard cells (Jezek & Blatt, 2017; Gray et al., 2020). Stomata characteristics generally study the shape of neighboring cells, type, number, density, length, and width of stomata in a leaf more deeply. The stomata distribution is closely related to the speed of the leaves' transpiration intensity, for example, the location of each other with a certain distance. Within a specific limit, the more pores, the faster evaporation. If the holes are too close together, then evaporation from one hole will hinder the evaporation of the nearby hole. The path taken by the water molecules through the hole is not straight but bends due to the influence of the guard cell's corners. The oval shape of the stomata makes it easier to expel water than the round shape (Folorunso, 2013; Shtein et al., 2017). Research on stomata characteristics of banana plant genomes AAB is useful for obtaining a stoma data base as an organ of gas exchange in the process of photosynthesis and respiration.

Method

This research was conducted at the Biotechnology Laboratory, Faculty of Mathematics and Natural Sciences, Pattimura University, Ambon, from January 3 to April 29, 2021. Tools used: binocular microscope (Olympus BX51) equipped with a camera (Olympus e-330), laptop, object-glass, pipette, cover glass, petri dish, digital camera, tweezers, razor, pin, and knife. The materials used were: plantain leaves and horn banana, Bayclin, 1% safranin, glycerin, label paper, tissue, and aquades. Procedure: Stomata observation was carried out by making an incision using the whole mount method (Damayanti, 2007). The scheme of the research method is shown in Figure 1.

The incision results were taken using an Olympus e-330 camera in a binocular microscope (Olympus BX51). The aim was to observe the shape of the guard cells, the type of stomata spread, the number of stomata, the number of neighboring cells so that the results are used to identify the type of stomata, the number of epidermal cells and measure the length, width, area, index and density of the stomata using the ImageRaster application which has been calibrated on a laptop. Data collection techniques were carried out quantitatively by counting the number of leaf stomata per stomatal field of view. Stomata density is calculated by the formula (Lestari, 2006):

$$\text{Stomata density} = \frac{\text{"Number of Stomata"}}{\text{"Stomata's Wide Field of View"}}$$

To measure the density of stomata, the field of view used at 40x magnification with the wide field of view was measured using the formula:

$$\text{Wide field of view} = \text{photo length } \times \text{photo width}$$

The distribution of stomata can be seen with parallel distribution and scattered distribution on one side of the leaf surface or both sides leaf surface (Haryanti, 2010). Therefore, the results were used to identify the type of stomata.

According to Lestari (2006), the observation of the stomata index on leaves is carried out with the following formula:

$$\text{Stomata index} = \frac{\text{"Number of Stomata"}}{\text{Number of Epidermal Cells + Number of Stomata"}}$$

The size of the stomata area in a cell was determined by measuring the length and width, and a sample was measured using the method (Franco, 1938) and (Wilkinson, 1979). Observation of stomata area on leaves was carried out with the following formula:

$$SS = \text{Stomata length x Stomata width x K}$$

Note: K= Franco constant, equal to 0.97.

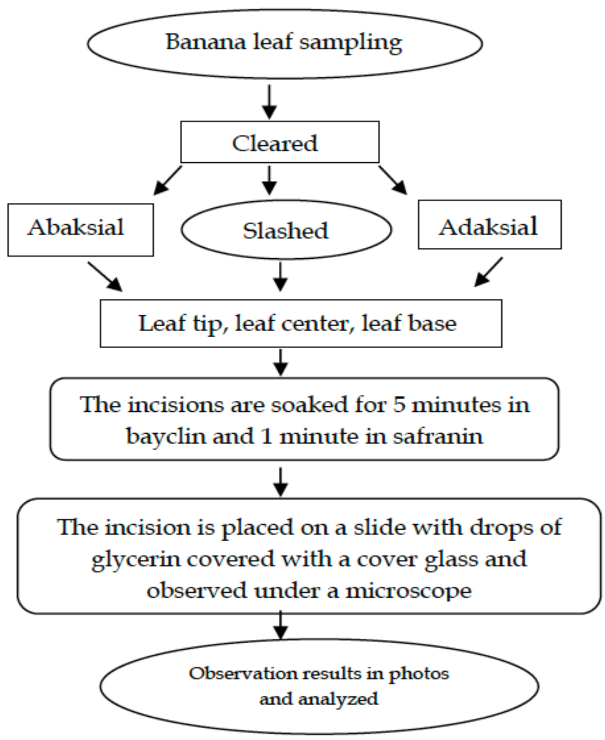

Figure 1. Schematic showing sampling and analysis of banana stomata

The results obtained were then analyzed descriptively qualitatively by describing the stomata characteristics of the plantain and horn banana plants.

Result and Discussion

The results showed that there were stomata on the upper and lower surfaces of plantain and horn banana leaves.

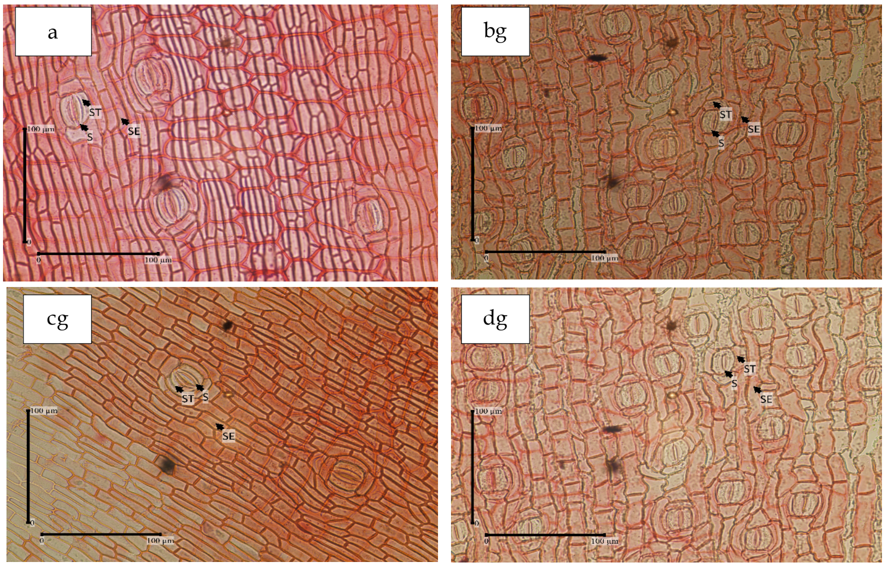

Figure 2. Stomata of banana leaves: (a) upper surface of plantain leaf; (b) Lower surface of plantain leaf; (c) the upper surface of the horn banana leaf; (d) the lower surface of the banana leaf. Description: S = stomata; ST= neighboring cell; SE = epidermal cells.

The results of the comparison of the stomata characteristics of the plantain and horn banana leaves can be seen in Table 1.

Table 1. The mean value of stomatal character variables/variables

Variable	Leaf Surface	Plantain	Horn Banana
Total	Adaxial	154.91	241.5
Epidermis	Abaxial	135	198.75
Number of	Adaxial	3.5	1.5
Stomata	Abaxial	13.91	18.33
Stomata	Adaxial	25.54	24.67
Length (µm)	Abaxial	21.67	22.11
Stomata Width	Adaxial	13.5	14.63
(µm)	Abaxial	11.33	13.85
Stomata Area	Adaxial	269.87	287.55
(µm^2)	Abaxial	194.65	245.95
Stomata Index	Adaxial	2.36	0.61
(%)	Abaxial	9.41	8.61
Stomata	Adaxial	43.75	18.75
Density (mm^2)	Abaxial	173.95	229.16

The observations showed that plantains and horn bananas have stomata in adaxial and abaxial locations. It shows that the type of distribution of stomata of plantain and horn banana based on the location of the leaf surface is amphistomatic, i.e., stomata are on both leaf surfaces (de Araujo et al., 2014). Plantains and horn bananas are monocotyledonous plants. The type of stomata is type 1 stomata, whose guard cells are surrounded by 4 to 6 neighboring cells (Stebbins & Khush, 1961).

The results of observing the characteristics of plantain and horn banana also show the average value of the number of the epidermis. For plantains in the adaxial section, the average value is 154.91. In the abaxial part of plantains, the average value for the number of the epidermis is 135. For horn banana in the adaxial section, the average value for the number of the epidermis is from horn banana with an average value of 241.5 and the abaxial part of the horn banana, the average value of the number of the epidermis is also higher than the plantain 198.75. It shows a number of the epidermis in the abaxial part of the plantain, and the horn banana is more than the adaxial part. However, when compared to plantains, the epidermis of the plantain is more.

For the number of stomata, plantain has an average value of the number of stomata on the adaxial part of 3.5 and banana horn 1.5. For the abaxial part, the average number of stomata for plantain is 13.91, and for horn, banana is 18.33. It shows that the number of stomata on the adaxial part of the plantain is higher than that of the horn banana. However, on the abaxial part, the number of stomata of the horn banana is higher than the plantain. This data shows that plantain and horn bananas also have more stomata on the abaxial part than on the adaxial. According to Harrison et al. (2020), this is influenced by plant physiological activities by avoiding stomata to direct sunlight so that stomata are more abundant on the underside of the leaves. According to Haryanti (2010), the number of stomata is grouped into categories: few (1-50), quite a lot (51-100), a lot (101-200), very lots (201-300), and infinity (301 - >700). Consequently, plantain and horn bananas are included in the low number of stomata because of their average value (1-50).

Based on Table 1, the length and width of the stomata on plantain and plantain leaves show a slight difference in comparing the average values. In the adaxial part, plantain has an average stomata length of 25.54 µm and stomata width of 13.5 µm. The adaxial part of the horn banana has an average length of 24.67 µm and an average value of stomatal width of 14.63 µm. In the abaxial part, plantain has an average stomata length of 21.67 µm and an average width of 11.33 µm. For the abaxial part, the banana horn has an average stomata length of 22.11 µm and an average width of 13.85 µm. The size of the stomata length refers to Juairiah (2014), which is grouped into less long (<20 µm), long (20-25 µm), and very long (>25 µm). According to Mustika et al. (2018), the width size is grouped into three categories, namely the less wide size (<19.42 µm), the wide size (19.42-38.84 µm), and the very wide size (>38.84 µm). Therefore, the plantain and horn banana both have stomata length, which is included in the long category, because of the average value of stomata length (20-25 µm) and for the width of stomata. Plantain and horn bananas are also included in the less wide size category because of the average value of the stomata width (<19.42 µm).

Based on Table 1, the stomata area of plantain in the adaxial area has an average value of 269.87. In the abaxial part, the average value of the stomata area is 194.65. For the horn banana in the adaxial section, the average value of the stomata area is 287.55, and the average value for the abaxial part is 245.95. This data shows that the difference of abaxial area is not far apart. However, the difference in the abaxial area of the stomata area of the horn banana is higher than the plantain.

Based on Table 1, the average value of the stomata index of plantain and horn banana compares the average values that are not too far apart. Plantain's adaxial surface has an average stomata index value of 2.36 µm and an average value of the index of the abaxial part of plantains 9.41. For the adaxial part of the banana, the average value of the stomata index is 0.61, and the abaxial part has the average stomatal index value of 8.61. According to Pompelli et al. (2010), a low number of stomata in a plant compared with a high number of epidermal cells will result in a low stomata index. Vice versa, if a high number of stomata is compared with a low number of epidermal cells, it will produce a high stomata index. If seen in Table 1, the stomata index of plantain and horn banana is low. It is because plantain and horn have a low number of stomata compared to the high number of epidermal cells. In table 1, it can also be seen that the average value of the stomata index of the plantain is higher than the horn banana. However, the number of stomata and epidermis of the plantain is higher. The range of the average value of the number of stomata and epidermis of plantains has a slight difference. However, for plantain, although the number of stomata and epidermis is high, the range of average values for the number of stomata and epidermis is very far. Therefore, the value of the stomata index of horn bananas is smaller than plantain.

It shows the average value of the stomata density of plantain and horn. In the adaxial part of plantain, the average value of stomata density is 43.75. In the abaxial part of plantain, the average value of stomata density is 173.95. For the adaxial part of the horn banana, the average value of stomata density is 18.75. For the abaxial part of the horn banana, the average value of the stomata density is 229.16.

This table also shows that the average value of stomata density in the abaxial part of the two bananas is higher than that in the adaxial part. This indicates that the number of stomata of these two bananas in the abaxial part is more than in the adaxial part. The high and low levels of stomatal density were seen based on categories according to (Rofiah 2010), namely low density (<300/mm^2), medium density (300-500/mm^2), and high density (>500/mm^2). The results showed that the level of stomata density on the adaxial and abaxial surfaces of plantain and horn banana was categorized as low stomatal density because of the average value of stomata density (<300/mm^2).

Conclusion

In conclusion, plantain and horn bananas have several characteristics in common, including kidney-shaped stomata, cover cells, the type of stomata spread is amphistomatic, the number of stomata on the lower surface of the leaf is more than the upper leaf, and the type of stomata on these two bananas is type one for monocot plants. The differences in the characteristics of plantain and horn bananas lie in the average value of the number of epidermises, number of stomata, stomata length, stomata width, stomata area, stomata index, and stomata density.

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