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Analysis of the Relationship between Snake Fruits Sidempuan (Salacca sumatrana Becc.) and Riring (Salacca zalacca var. amboinensis) using the Morphological Characterization Approach

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KEYWORDS

diversity;
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morphological
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Abstract Genetic relations estimation can be used for germplasm management activities, identifying cultivars, selecting parents for crossing, and reducing the number of individual samplings in a wide range of genetic diversity. The research's fundamental purpose is to analyze kinship relationships and study the genetic diversity between the snake fruit Sidempuan plant from South Tapanuli with snake fruit Riring plants from Ambon Island. Characterization research was conducted in Padang Sidempuan, South Tapanuli, and Riring Village, Ambon Island, using Radford characterization guidelines. The morphological characterization results showed that the Sidempuan and Riring bark's ecotypes were divided into 3 main clusters and had a kinship relationship with the value of genetic similarity of 85% - 100%. Even though it has similarities, there were several distinguishing characters, namely the color of the upper surface of the leaf, the folding of the edges of the leaf blade, the taste of the flesh of the fruit, the height of the plant, and the category of snake fruit flowers. In a nutshell, although they looked similar, the two ecotypes were not identical.

Introduction

Snake fruit (*Salacca zalacca*) is one of the fruit plants that originated from Southeast Asia (Indochina, Malaysia, Thailand, and Indonesia) (Mogea 1990, *in* Ashari, 2013). The cultivation of this plant is continuously developing in Indonesia because of the increasing market demand and its high economic value (Sudjijo, 2009) as well as its lofty nutritional content (Nandariyah, 2004).

There are two places in Indonesia known as the snake fruit producing centres, such as Padang Sidempuan, South Tapanuli, North Sumatra, and Riring Village, Ambon Island. There are two kinds of snake fruit fruit in both locations based on the color of the flesh, which is white and red. This variation is perhaps how this plant propagated, i.e., by sexual method (using seed) (Suskendriyati *et al.*, 2000; Nandariyah, 2004). However, the research results in reviewing the genetic diversity of the snake fruit plant are still limited. It is further compounded by giving names that are still according to the origin of the location where the cultivation is (Ashari, 2013).

Nandariyah (2008) stated that the snake fruit breeding program was carried out to obtain new superior varieties by utilizing the genetic diversity of the snake fruit plant in the still lacking regions. Besides, the unavailability of germplasm as a source of genes in plant breeding programs also impacts the genetic content of the varieties produced. In other words, it will uniform the background of germplasm of a variety (Sumarno & Zuraida, 2008). The variation of fruit flesh of snake fruit in North Sumatra and Ambon island (white and

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red) color is needed to be investigated, and the relationship of both types is necessary.

Similarities and differences in the morphological characteristics of a plant can also be used to determine the relationship among them (Suskendriyati *et al*., 2000). Moreover, Santos *et al*. (2011) stated that observations of morphological characters are done by directly observing plants' characteristics visually with ordinary eyes in the same environmental conditions. Scientific studies on the use of morphological characterization methods have been carried out by several researchers for plants such as snake fruit plants (Harahap *et al*., 2013; Sudjijo, 2009) and bananas (Radiya, 2013). This study aimed to study and analyze the relationships and genetic diversity of Snake Fruit Sidempuan from South Tapanuli, North Sumatra, and Snake fruit Riring from Ambon Island.

Materials and Methods

The morphology characterization study of snake fruit plants was conducted from April to May 2017 in two different locations, which became the centers of snake fruit plant production. The characterization activity began with a survey of the gardens of local farmers. The main ingredients used for morphological characterization were snake fruit plants in the city of Padang Sidempuan and Riring village. The tool used in the study was a guide to the characterization of snake fruit plants, according to Radford (1986), writing instruments, rulers, marker tapes, and digital cameras.

Data from morphological characterization observations were presented descriptively (Nandariyah, 2008). The data was then converted into binary data, and if there were properties that do not appear with properties compared to zero (0). Instead of given a value of one (1) if it showed the properties that appear, then cluster analysis was carried out and displayed in the form of a dendrogram using the Unweighted Pair - Group Method With Arithmetic (UPGMA) method through a multivariate PAST (Paleontological Statistics) program.

Results and Discussion

Morphological Characteristics of Snake Fruit Sidempuan and Snake Fruit Riring

The result of the investigation by the morphological method is presented in Table 1.

Table 1. Morphological Characteristics of Snake Fruit Sidempuan (*Salacca sumatrana* Becc.) Snake Fruit Riring (*Salacca zalacca var. amboinensis*)

The characteristics of snake fruit plants were observed	Ecotypes of snake fruit	
	Snake Fruit Sidempuan	Snake Fruit Riring
Vanished color	Brown	Brown
The surface color of the leaf	Dark green	Shiny green
The color of the bottom surface of the leaf	Grayish green	Grayish green
Wax coating thickness	Thin	Thin
Midrib color	Green	Green
Number of leaflets	72-88	69-70
Distance between leaflets (cm)	2.5 – 6.5	3 – 4
Leaf hardness	Hard	Hard

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The characteristics of snake fruit plants were observed	Ecotypes of snake fruit	
	Snake Fruit Sidempuan	Snake Fruit Riring
The shape of the base of the leaflets	Uneven	Uneven
The shape of the tip of a leaflets	Pointed	pointed
Folding the edge of the strand	Present	Not present
Midrib thorns color	Black/brown	Black
Sharpness of thorns	Sharp	Sharp
Hardness of thorns	Hard	Hard
Easy thorns off	Not present	There is no
Thorn shape	Thin, pointed, small	Thin, pointed, small
Density of thorn	Rather density until density	Density
The color of the flower sheath	Brown	Brown
The shape of a flower sheath	Lengthwise	Lengthwise
Flower crown color	pink, yellow red and dark red	pink, yellow red and dark red
Number of fruit each bunch	16-35	15
Ripe fruit skin color	brown, yellow-brown, and blackish-brown	Brown and yellow-brown
Fruit shape	short triangle and long triangle	short triangle
Color of fruit flesh	white, a tinge of red, white, dark red, and a tinge of yellow	White and red
The taste of fruit flesh	Sweet, sweet-sour taste and bittersweet taste	Sweet and sweet-sour taste
Texture of fruit flesh	Mushy texture (soft and juicy)	Mushy texture (soft and juicy)

The similarity of several morphological characters between the two ecotypes of snake fruit plants in these two locations did not mean that these two ecotypes were identical or unidentical so that these differences could be used for taxonomic activities (Jones *et al.*, 1986). According to Sitompul and Guritno (1995), these similarities and differences were influenced by environmental factors and genetic factors. They explain the basic theory of genetic science, which states that the interaction between environment and genotype will give rise to a phenotype.

The character that distinguishes the feature of the Padang Sidempuan with Riring bark based on the Radford's characterization guide (1986) is on the color of the upper surface of the leaf, folding the edges of the leaf blade, and the taste of fruit flesh. Snake fruit is categorized as good quality and customer favorite if the fruit meat does not have bittersweet (Sudjijo, 2009). It is in line with Anas (2004) that snake fruit with a large size, sweet fruit taste, thick fruit flesh, and perfectly teardrop shape is the fruit features that appeal to consumers and are highly demanded. Related to the number and size of snake fruit fruit which

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varied both on the ecotypes of Snake Fruit Sidempuan fruit and Riring bark, they were allegedly influenced by the success of pollination and agronomic actions, and the level of fruit density in the cluster (Baswarsiati *et al.*, 1993; Nandariyah *et al.,* 2000).

Another distinguishing feature found in the ecotypes of snake fruit plants originating from Padang Sidempuan and Riring is the character of plant height and pollination mechanism. From the results of the study, it was known that the Snake Fruit Sidempuan plant had plant height ranging from 6.5-7 meters. The size of the Snake Fruit Sidempuan tree is large and has a wide canopy. It is a cross-pollinated plant and is classified as dioecious. While the height of snake fruit Riring plants ranges from 5-6 meters, it is a self-pollinating plant and includes hermaphrodite flowers.

Site height factors can also influence snake fruit plants' leaf color and different plant height between ecotypes in both study locations. According to the survey, it is known that the research location in Padang Sidempuan is at an altitude of 400-600 m above sea level, while Riring Village is at an altitude of about 600 m above sea level. Areas with high altitude will affect the intensity of sunlight and air temperature. Besides, CO_2 concentrations in areas with high low places are greater than areas with high altitudes. In such conditions, this will directly affect the process of photosynthesis. The number of leaves looks almost the same in the two locations studied because, according to Rachman *et al*. (1991), the character of the number of leaves has a high heritability so that climatic conditions, including the growing environment, do not influence this character.

The characterization of snake fruit Sidempuan and snake fruit Riring also showed a red color in the flesh but with varying color classification. Snake Fruit Sidempuanin Padang Sidempuan has red color variations such as red, small points, tinge, dominant tinge, and total red. In contrast, the red color of snake fruit plant in Ambon is the only tinge as it is known that the red color is due to anthocyanin content.

According to Delgado-Vargas *et al*. (2000), the anthocyanin synthesis process in plant bodies is very synergistic with environmental conditions such as temperature, nutrient composition in the soil, rainfall, and sunlight intensity. The cold conditions environment is very supportive of anthocyanin synthesis (Kim *et al*., 2014). In the meantime, Brown *et al*. (2008) stated that the higher the land, the more anthocyanin content is produced in the plant's body.

In addition to environmental influences, an individual's qualitative character's appearance is also controlled by genes or interactions between the two. In the body of plant organisms, many genes control various types of properties. Each gene has a special function to regulate an individual characteristic. The character can be in the form of morphological characters such as size (height, length, and width), color, shape, and habitus (Maruapey, 2012).

Relationship Between Several Ecotypes of Snake Fruit Plant

Qualitative data of several snake fruit ecotypes obtained from the subsequent characterization results are displayed in the form of dendrograms, as shown in Figure 1.

The results of genetic similarity analysis based on morphological character data from 20 ecotypes of snake fruit plants tested were 0.85-1.00 (85-100%) with a percentage of the diversity of only 15% and 0%. The dendrogram also showed the existence of 3 main clusters formed (Figure 1). The first ecotype of snake fruit included in the cluster is the red and white ecotype of Snake Fruit Riring. Both ecotypes depicted similarities in the range of similarity values of 0.86 or 86%. The second group consisted of several ecotypes, including Snake

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Fruit Padang Sidempuan fruit of yellow tinge$_2$, Snake Fruit Padang Sidempuan fruit of red$_2$, Snake Fruit Padang Sidempuan fruit of red$_5$, Snake Fruit Padang Sidempuan fruit of tinge$_5$, Snake Fruit Padang Sidempuan fruit of red$_3$, and Snake Fruit Padang Sidempuan fruit of red$_4$. (same subscript indicate the same red colour rank). In comparison, the third cluster was the ecotypes of the Snake Fruit Padang Sidempuan fruit of white, tinge (except Snake Fruit Padang Sidempuan fruit in tinge$_5$), Snake Fruit Padang Sidempuan fruit of red$_1$ and Snake Fruit Padang Sidempuan fruit of yellow tinge$_1$. Nevertheless, there are also some 100% similar ecotypes, among others of Snake Fruit Padang Sidempuan fruit of tinge$_1$ with Snake Fruit Padang Sidempuan fruit of tinge$_2$, also Snake Fruit Padang Sidempuan fruit of tinge$_3$ with Snake Fruit Padang Sidempuan fruit of yellow tinge$_1$.

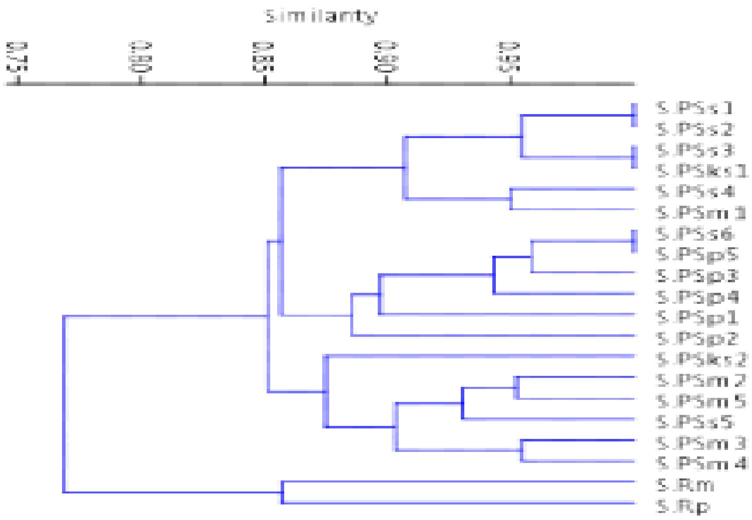

Figure 1. Dendrogram of qualitative data characterization of eco typical morphology of snake fruit from Padang Sidempuan and Riring

The other sub cluster showed that Snake Fruit Padang Sidempuan fruit of tinge$_1$, Snake Fruit Padang Sidempuan fruit of tinge$_2$, Snake Fruit Padang Sidempuan fruit of tinge$_3$ and Snake Fruit Padang Sidempuan fruit of yellow tinge$_1$ had similarities with snake fruit Padang Sidempuan fruit of tinge$_4$. Meanwhile, Snake Fruit Padang Sidempuan fruit of tinge$_1$, Snake Fruit Padang Sidempuan fruit of tinge$_2$, Snake Fruit Padang Sidempuan fruit of tinge$_3$, Snake Fruit Padang Sidempuan fruit yellow tinge$_1$ and snake fruit Padang Sidempuan fruit tinge$_4$ had similarities to the Snake Fruit Padang Sidempuan fruit yellow tinge$_2$ in the range of 0.88 (88%). The high morphological similarity value of 0.97 or 97% was also shown by Snake Fruit Padang Sidempuan fruit of tinge$_6$ and Snake Fruit Padang Sidempuan fruit of white$_5$ with Snake Fruit Padang Sidempuan fruit of white$_3$.

Based on the study results, which showed that the percentage of genetic similarity between the ecotypes of Padang Sidempuan and the Riring ecotype is 85%, it means that these two ecotypes have a narrow genetic diversity of 15%. Considering a close kinship relationship, this will certainly be difficult for character improvement if a crossing is carried out between the two ecotypes. In line with the results, Cahyarini *et al*. (2004) delineated that a kinship relationship is accounted to be far if it has a similarity value of less than 0.60 or 60%. According to Nei (1987) and Hardiyanto *et al*. (2008), genetic similarity is the opposite of genetic distance. The smaller the similarity coefficient value, the further the genetic relationship between individual plants. The high level of morphological similarity or genetic chemistry of a plant indicates that plants' genetic diversity is getting lower and vice versa.

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A similar number of characters can indicate that among individuals or populations have a kinship relationship, assuming that genetic makeup differences cause different characters.

In the science of plant breeding, information about descriptions, kinship, and genetic distance is crucial for the determination and selection of elders. The far genetic distance between prospective crossing parents, the better chance of producing hybrid plants has a low compatibility and fertility level. It would be better if the crossing is carried out between closely related elders so that it will produce a new plant that not only has narrow genetic diversity but also has a high fertility rate (Hadiati *et al*., 2009; Yullianida *et al*., 2016; Collard and Mackill, 2008). Therefore, high or wide genetic diversity is a supporting factor for the success of breeding programs.

Conclusions and Suggestion

Based on this research, we assume that the ecotypes of snake fruit plants originating from Padang Sidempuan and Riring were grouped in 3 main clusters and had close kinship relationships with genetic similarity values of 85% - 100%. Although the two ecotypes have similarities, there are several distinguishing characters such as color of the upper surface of the leaf, the folding of the edges of the leaves, the taste of the flesh of the fruit, the plant's height, and the category of snake fruit flowers. Then, we suggest that further research needs to be done using specific primers for snake fruit plants to confirm the relationship and genetic diversity from a more accurate molecular perspective.

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