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Phenetic Taxonomy of Local Duku (*Lansium parasiticum* (Osbeck) K.C. Sahni & Bennet) from Three Regencies Based on Micromorphological Characters

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Abstract. *Lansium parasiticum* (Duku) is a plant species with complex morphological characters because of its wide distribution. This study examined the plant anatomical variation patterns and phenetic relationships among 15 local duku samples from Purwokerto, Purbalingga, and Banjarnegara. The anatomical data were analyzed descriptively, and the similarity relationship was measured with the UPGMA method using the MEGA 10.0.1 software. The results of anatomical characters analysis suggested a high degree of diversity among the duku. UPGMA tree derived from cluster analysis showed two majors clusters. The first cluster consisted of samples whose upper epidermis thickness was $\geq 16.5\mu\text{m}$. The second cluster comprised fourteen samples with upper epidermis thickness $< 16\mu\text{m}$.

1. Introduction

Locally known as Duku (*Lansium parasiticum*), this plant is a native to the Southeast Asian region and is one of Indonesian seasonal popular fruit. It has several names as luku, kokosan, and langsung in Indonesian; duku and langsung in Burmese; langsung and duku in English; lanzone, lanzon, lansones, and lanson in the Philippines; langseh, langsep and elderly in Malaysia; duku, longkong, langsung in Thailand; bonbon in Vietnamese [1]. The plant is tropical fruit which economically valuable and containing necessary nutrition. In Indonesia, it naturally distributes across Sumatra, Kalimantan, Sulawesi, and Java [2]. In some parts of those regions, Duku is planted and cultivated.

Duku has several cultivars and the most famous cultivar in Indonesia is Palembang. In Java, it is Condet, but formerly it included Menteng and Depok cultivars from around Jakarta, Papongan from Tegal, Kalikajar from Purbalingga, Karangajen and Klaten cultivars from Yogyakarta, Matesih from Karanganyar, Woro from Rembang and Kudus, and some others. In South Kalimantan, it was the Padang Batung cultivar from the Hulu Sungai Selatan Regency [3, 4].

Duku variety is a population consisting of one or several biotypes. It has different morphological characteristics, distributed locally in a limited area within the species distribution ranges, so the varieties are referred to as local race [5, 6]. The occurrence of morphological variations of the trees, leaves, and fruits has caused different identifying names in several regions [7]. The number of varieties or cultivars leads to difficulties in distinguishing Duku. Therefore a reliable scientific grouping of varieties or cultivars using numerical taxonomy methods is urgently needed. The approach uses morphological, anatomical, chemical, cytological, isozyme, or DNA features [8]. The standard identification and characterization of plants varieties to date is not only based on morphology but also anatomy and physiology [9]. Micromorphology or anatomy of leaf epidermises such as cuticles,

stomata, and trichomes has been confirmed as the most useful characteristic to distinguish species in taxonomic studies [10]. Anatomical features also essential to solve complex plant taxonomic difficulties in identification at the family, genus, or species level. It will then supports the accuracy of plant identification to which it is crucial for plant breeders, ecologists, and conservationist [11].

2. Methods

Observation of the anatomy of the Duku leaf was started with the preparation for the transverse cutting using the paraffin method referring to Sass [12]. The observed characters included the number of stomata, cuticles, epidermis, palisade ratio, and trichomes. Data were calculated using the Unweighted Pair Group Method with Arithmetic mean (UPGMA) cluster analysis performed in the MEGA 10.0.1 software.

3. Results

Table 1. Anatomical characters

	TM	TEPM	RP	JTA	LTA	
Purwokerto 1	163	12.5	11.6	2.6	21	
Purwokerto 2	228	10.5	11.8	3	20.5	
Purwokerto 3	188.5	9	13.6	3.2	22.5	
Purwokerto 4	204	9.5	13.4	2.6	20.5	
Purwokerto 5	215.5	8.5	9.8	2.2	20	
Purbalingga 1	191.5	8	11.6	2	19	
Purbalingga 2	228.5	6.5	11.4	2.4	19.5	
Purbalingga 3	162	7	11.4	2.2	19	
Purbalingga 4	185.5	6.5	9.2	2.6	18.5	
Purbalingga 5	167.5	6.5	10.8	2.8	22	
Banjarnegara 1	174.5	11	9.2	3.2	18	
Banjarnegara 2	154	8	11.2	3.2	19.5	
Banjarnegara 3	173	10.5	11.6	2	19.5	
Banjarnegara 4	177.5	7.5	9.8	3.6	20	
Banjarnegara 5	150.5	9.5	10.2	3	17	
	LTB	JS	PS	LS	TK	TEA
Purwokerto 1	19	10	16	2.5	6	14
Purwokerto 2	21	11	16	2.5	7	14
Purwokerto 3	19	7.8	16	3	7.5	12.5
Purwokerto 4	19	9	18	3	6	15.5
Purwokerto 5	19	8.8	16.5	2.5	5.5	18
Purbalingga 1	21.5	10.6	15	3.5	3.5	13
Purbalingga 2	21.5	10.2	15	3.5	3	12
Purbalingga 3	20	9.4	17.5	3.5	4	12
Purbalingga 4	19	10.6	15.5	3	4.5	12
Purbalingga 5	21	7.6	14.5	3.5	3.5	10.5
Banjarnegara 1	21.5	10	21.5	3.5	5.5	13.5
Banjarnegara 2	19.5	11	19.5	3.5	4	13
Banjarnegara 3	19.5	13	19.5	3.5	5	16.5
Banjarnegara 4	21	10	21	4	3.5	11
Banjarnegara 5	21	10.2	21	3.5	4	13.5

TM	: mesophyll thickness	TEPM	: lower epidermis thickness
RP	: palisade ratio	JTA	: upper trichome number
LTA	: upper trichome width	JTB	: lower trichome number
LTB	: lower trichome width	JS	: stoma number
LS	: stoma width	PS	: stoma length
TK	: cuticle width	TEA	: upper epidermis thickness

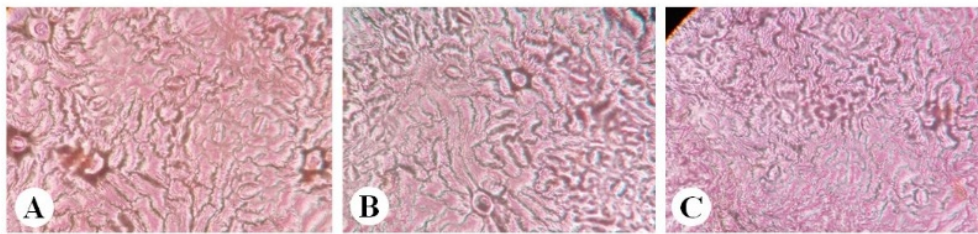


Figure 1. The stoma anatomical structure of Duku leaves (*LansiumParasiticum* (Osbeck) Sahni & Bennet) A. Banjarnegara, B. Purwokerto, C Purbalingga . The stoma type is anomocytic

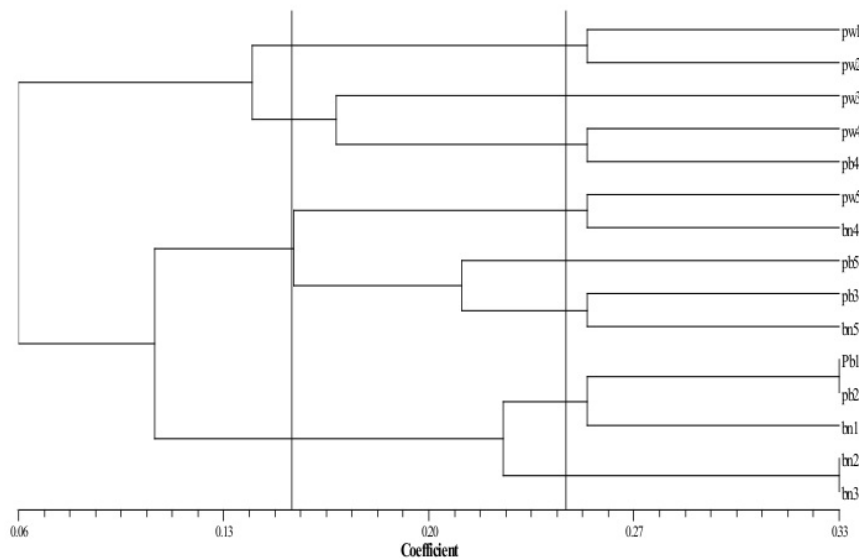


Figure 2. Similarity Based on Anatomical Characters

4. Discussion

The anatomy of the leaf cross-section consists of several tissues, the upper epidermis, mesophyll, and lower epidermis [13]. Epidermal tissue is a collection of cells that are uniform and are in the outer layer. Epidermal cells have a solid compact structure with cell walls sometimes thickened because of the silica content, thus strengthening the leaf structure. In general, the epidermal tissue hold stomata, trichomes, and other features. Stomata that function in gas exchange between leaf tissue and the atmosphere sometimes were found on the surface of the upper or lower leaf or both.

The results of this study recognized the anatomical characteristics of the stomata were found only on the underside of the leaf or abaxial, 25 stomata types [14]. Stomata types are distinguished based on neighboring cells surrounding the guard cells in the stomata [15]. Duku stomata from 3 regions have the same stoma type, anomocytic type. The shape is the kidney-shaped stomata covering cells, scattered irregularly on the leaf surface of the abaxial part. In most species, the frequency of stomata in the lower epidermis is more than the upper epidermis [16]. The distribution of stomata mostly influenced by internal relations and external organs [17]. The average number of stomata of Duku leaves from Purwokerto, Puralingga, and Bajrangewara were 8-11 stomata/mm², 8-10 stomata/mm², and 10-13 stomata/mm², respectively.

The observed size of stomata in Duku leaves varied with lengths varying from 15-21 μm and widths from 2.5 to 4 μm (Figure 1). The largest stomata were found in Banjarnegara Duku with a measured height of 21 μm and 4 μm wide. The thickening of guard cells influences variations in the size of the stomata in response to light, CO_2 , and water content [18].

The epidermis is the outermost plant tissue that generally consists of only a single cell or cells, which functions to protect the internal tissues. In the leaves, the epidermis also serves to reduce transpiration. Therefore it is often covered with cuticles and water-resistant waxes [19]. Based on the observations from the three districts, the epidermis thickness of the Duku leaves was 17 μm - 21 μm . The Duku leaves having the thickest layer of the upper epidermis were of Purwokerto with 21 μm thick. The lower epidermis thickness varied from 7 to 12.5 μm . The Duku from Purwokerto has the thickest of the lower epidermis, which measured 12.5 μm . The thickness of leaf tissue layers such as abaxial epidermis, adaxial epidermis, mesophyll, and palisade showed significant variations among all accessions examined. The differences in leaf layer thickness may be related to responses to environmental factors [20].

The Leaf mesophyll is a tissue that occupies beneath the epidermis layer, which usually is differentiated into photosynthetic tissue containing chlorophyll [19]. The observation of transverse cutting anatomical structure showed that the mesophyll tissue was separated into palisade and spongy parenchyma. Palisade network is a network arranged tightly between cells, while spongy tissue has a lot of space between cells compared to the palisade network [21]. The Palisade layer of Duku leaves is a bifacial property that only found on one adaxial side. Palisade network in each species has a different number of cell layers.

The numerical analysis presents a phenetic relationship between 15 Duku cultivars. The phenograms constructed based on cluster analysis, revealed two clusters, the first cluster consisting of five cultivars PW1, PW2, PW3, PW4, and PB4. The second cluster consisted of one cultivar containing 11 cultivars. The grouping does not always indicate a similar geographical origin but may show genetic similarity [22].

The results of the dendrogram relationship can be used as a reference for determining the parent in making seedlings. The farther the connection between individuals or the smaller the genetic distance, the smaller the success of the crosses, but the possibility of obtaining superior genotypes is more significant if the crossbreeds are successful. The farther the relationship, the more genetic variety each sample carries, the more likely a superior genotype is obtained. If the hybridization is carried out between close genetic individuals or the same connection has the effect of increasing homozygosity. Conversely, the hybridization between individuals with considerable genetic distance or distant relationship has the effect of increasing heterozygosity. This information has a good impact on the process of making superior seeds. The breeding of an ancestor with a relatively high genetic variation will produce individuals with higher heterozygosity [23].

5. Conclusion

The anatomical features have successfully distinguished Duku cultivars used in this study. The leaf characters used in this study can be used as a diagnostic tool for plant identification.

6. Acknowledgement

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