

Morphological Characterization of 10 Clones of Superior Sugarcane (*Saccharum officinarum* L.) Mutants Resulting from Colchicine Induction at PT Gunung Madu Plantations

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Abstract: Sugarcane (*Saccharum officinarum* L.) is a sugar-producing plant commonly cultivated in tropical climates. However, frequent decline in its productivity often results in unmet sugar demand. Sugarcane breeding techniques are thus required to increase productivity using the chemical mutagen colchicine. This study aims to evaluate the diversity of morphological traits of 10 superior mutant sugarcane clones induced by colchicine, derived from the varieties GMP6, RGM047, RGM1183, and RGM186. It was conducted at PT Gunung Madu Plantations using a randomized block design. Experimental units were grouped according to their original variety GMP6 (clones 36, 44, and 63), RGM047 (clones 160 and 162), RGM1183 (clones 200, 202, and 226), and RGM186 (clones 293 and 296), with 3 replicates. The parameters observed included germination grading, ground cover, leaves, and leaf midribs. Data analysis was performed using the Multivariate Statistics Package (MVSP) version 3.2, and genetic distance was used in cluster analysis using the Unweighted Pair Group Method with Arithmetic Mean (UPGMA). The results indicated significant differences in germination, ground coverage, leaf, and frond characteristics. Cluster analysis revealed a relatively close phenetic relationship between the original varieties and their superior mutant clones. These results suggest that colchicine induction effectively generates genetic variation among the 10 superior mutant sugarcane clones.

Keywords: Colchicine, morphology, relationships, sugarcane, superior clones.

1. Introduction

Sugarcane (*S. officinarum* L.) is one of the primary sugar-producing plants (Windiyani et al., 2022). According to data from the Food and Agriculture Organization (FAO) (2020), sugarcane productivity in Indonesia has declined to 21.74 million tons since 2018. This figure is lower than in other sugar-producing countries, such as Brazil (746.83 million tons), India (376.9 million tons), China (108.72 million tons), and Thailand (104.36 million tons). The primary factors contributing to low sugarcane production in Indonesia are reduced sugarcane area (Aritonang et al., 2024), plant pests and diseases (Mahfut, 2020; 2021; Mahfut et al., 2020a; 2020b; 2020c; 2021b; Anbiya et al., 2024; Septiana et al., 2024), and the unavailability of superior seeds that are adaptive to environmental conditions (Sukmawati et al., 2021), leading to decreased productivity (Putera et al., 2024).

PT Gunung Madu Plantations (GMP) is a pioneer in the sugar industry in the Lampung region, Indonesia, and implements a superior sugarcane cultivar breeding program. Several superior varieties of PT GMP, namely GMP3, GMP6, RGM047, RGM1183, and RGM186, have been widely reported to be cultivated due to their large population and drought resistance. However, these varieties exhibit low-quality morphological characteristics, such

as short plant stature, few leaves, and narrow leaf width (Windiyani et al., 2022; Mahfut et al., 2023a). Increasing sugarcane productivity can be achieved by ensuring the availability of superior seeds. One such approach involves the use of mutation techniques, which can use physical, chemical, and biological mutagens to develop new superior plants through genetic material variation (Asadudin et al., 2024).

A commonly used mutation technique involves the chemical mutagen colchicine ($C_{22}H_{25}O_6N$), a white alkaloid derived from the seed extract of *Colchicum autumnale* L. This compound is known for its ability to induce chromosome duplications in plants (Nuraini et al., 2024). Chromosome duplication leads to an increase in the number and variety of alleles, which are determined by the chromosomes. Alleles represent gene variations that influence traits in plants. Consequently, the duplication of chromosomes enhances genetic diversity and can lead to improved productivity in plants (Mahfut et al., 2025a; 2025b; 2025c).

Mahfut et al. (2021a) explained that in addition to agronomic characteristics (Mahfut et al., 2024a), anatomical characteristics (Asadudin et al., 2024; Putera et al., 2024), and molecular characteristics (Sari et al., 2024; 2025), morphological characteristics are important aspects that need to be evaluated in plant breeding induced by colchicine mutation, which is conducted through the observation of sugarcane growth (Putra et al., 2024a; 2024b). Several studies on the morphological

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characteristics of colchicine-induced sugarcane at PT GMP have been conducted on various plant organs, including leaves, stems, internodes, and buds. Mahfut et al. (2024a) demonstrated morphological diversity in the 21 mutants of the GMP3 variety, which exhibit medium-sized leaf width, dark green leaf color, branched leaf shape, absence of dorsal hairs, cylindrical internode shape, medium internode length (>13 cm), and medium-sized stem diameter (2.5–3 cm).

The study on the morphological characteristics of colchicine-induced mutant sugarcane in the GMP6, GM-047, GM-1183, and GM-186 varieties continues previous research (Nuraini et al., 2024; Simamora et al., 2024). Nuraini et al. (2024) reported observations of mutant morphological characteristics in 5 sugarcane varieties, namely GM-120, GM-86, and GM-25, at 0 to 1 month after application, which exhibited stable germination, indicating the formation of plant organs at each stage without significant growth inhibition. Meanwhile, the GM-10 variety had slower germination than the other varieties, especially in the formation of leaf organs and shoot roots. Simamora et al. (2024) also reported significant morphological changes in plant height, number of leaves, and leaf area of 10 selected superior mutant clones of the GMP6, GM-047, GM-1183, and GM-186 varieties at 3 months of age. Therefore, further research is needed on the morphological characterization of 10 superior colchicine-induced mutant sugarcane clones at 7 months of age. This study aims to determine the stability of the morphological characteristics of the 10 superior mutant clones when planted in agricultural fields.

2. Methods

Plant Material

Morphological character observations were carried out directly on the control and 10 superior mutants of the GMP6, RGM047, RGM1183, and RGM186 sugarcane variety induced by colchicine with concentrations of 0.1% and 0.2%, aged seven months at PT Gunung Madu Plantations.

Morphological Trait Observation and Analysis

Observation of morphological characteristics followed the procedures established by PT GMP. The observation parameters used were scoring germination, scoring ground coverage, leaf morphological characters, and leaf midrib morphological characters. The scoring system used a rating of 1 to 5, where 1 = very little, 2 = a little, 3 = medium, 4 = a lot, and 5 = very much. The method used in this research was the observation method with a Randomized Block Design (RBD) on original varieties and superior mutant clones. Data analysis was carried out following the methods of Mahfut et al. (2024a; 2024b), which involved describing the results of observations and then phenetic analysis using the Multivariate Statistics Package (MVSP) version 3.2 and genetic distance in cluster analysis using Unweighted Pair Group Method with Arithmetic Mean (UPGMA). Next, to determine the morphological characters that influence the grouping between species, we used Principal Component Analysis (PCA).

3. Research Results

The study samples used were from four control types and their superior mutant clones, namely GMP6 (clones 36, 44, and 63), RGM047 (clones 160 and 162), RGM1183 (clones 200, 202, and 226), and RGM186 (clones 293 and 296). Germination grading observations were carried out using scoring or direct evaluation in the field, with clone scores of 4 to 5 indicating the influence of colchicine on the sample, which results in a large number of germinations in sugarcane. The germination grading scores are presented in Table 1.

Table 1. Scoring Grading Germination on 10 Superior Mutant Sugarcane Clones at PT Gunung Madu Plantations

Sample	Scoring
GMP6	3
Clone 36	3
Clone 44	2
Clone 63	3
RGM047	3
Clone 160	2
Clone 162	1
RGM1183	3
Clone 200	4
Clone 202	2
Clone 226	4
RGM186	4
Clone 293	2
Clone 296	3

The germination grading results observed in 7-month-old sugarcane showed variations across the samples. The application of colchicine to mutant clones influenced germination grading, although not uniformly across all superior mutant clones. The test sample scoring results of superior varieties and mutant clones with the highest germination were clone 200, clone 226, and RGM186. This indicates the influence of colchicine on clone 200 and clone 226. Administering colchicine can stimulate germination growth, resulting in an increase in the number of germinations in the sample.

The use of colchicine increases plant germination and growth compared to the control, but this depends on the concentration applied (Sadiq et al., 2025). This statement is supported by Lv et al. (2021), who reported decreased germination percentage due to increasing concentration. Therefore, colchicine had significant effect on any of the mutant clones. Increased colchicine concentrations may also result in a low germination percentage. This is because colchicine can inhibit cell division. Germination therefore slows down because the cells cannot divide quickly (Mahfut et al., 2023b). As a result, the vegetative phase is also a phase of cell growth and development.

The next parameter to consider is the ground cover. Ground coverage is the percentage of land area covered by sugarcane plants and is used to assess the shape of leaves or curved sheets on sugarcane leaves that can protect them from surrounding weeds (Pringgani et al., 2023). Land cover observation was carried

out using scoring or direct evaluation in the field, with evaluation results between 1 and 5, with the information that 1 = very low, 2 = low, 3 = medium, 4 = high, and 5 = very high. This scoring

assessment is carried out qualitatively by examining the strands of sugarcane leaves covering the ground. The land coverage scores are presented in Table 2.

Table 2. Scoring Ground Coverage on 10 Superior Mutant Sugarcane Clones at PT Gunung Madu Plantations

Sample	Scoring
GMP6	3
Clone 36	3
Clone 44	3
Clone 63	3
RGM047	3
Clone 160	3
Clone 162	3
RGM1183	4
Clone 200	3
Clone 202	4
Clone 226	3
RGM186	4
Clone 293	3
Clone 296	3

The scoring results of the test samples for superior varieties and mutant clones were RGM1183, clone 202, and RGM186, which had the highest ground coverage among the samples with a score of 4. The effect of colchicine was evident in clone 202 of the variety RGM1183. According to Yuniyati et al. (2022), colchicine induction can affect the characteristics of pulsative mutant leaves, one of which is the curvature of the leaf. The results of observations in this research show that clone 202 has better soil

coverage. This is shown by the number of leaves in the sample row of clone 202 that cover the surface of the soil, further causing weeds not to grow in the planting area of clone 202. The third parameter is the leaf character. The results of leaf morphological characteristics observations of GMP6, RGM047, RGM1183, and RGM186 and superior mutant clones at PT Gunung Madu Plantations are presented in Tables 3-6.

Table 3. Leaf Morphological Characteristics of GMP6 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Character	GMP6	Clone 36	Clone 44	Clone 63
Curved Leaf Blade	<1/3 Leaves	<1/3 Leaves	<1/3 Leaves	<1/3 Leaves
Leaf Width	Narrow (<4cm)	Narrow (<4cm)	Narrow (<4 cm)	Narrow (<4 cm)
Leaf Color	Yellowish Green	Yellowish Green	Yellowish Green	Yellowish Green
Leaf Triangle Color	Purple	Purple	Purple	Green
Leaf Ears	Weak	Earless	Earless	Weak
Position of Ear Leaves	Upright	Upright	Upright	Upright

Table 4. Leaf Morphological Characteristics of RGM047 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Character	RGM047	Clone 160	Clone 162
Curved Leaf Blade	<1/3 Leaves	<1/3 Leaves	<1/3 Leaves
Leaf Width	Narrow (<4cm)	Narrow (<4cm)	Narrow (<4cm)
Leaf Color	Yellowish Green	Yellowish Green	Green
Leaf Triangle Color	Green	Green	Green
Leaf Ears	Earless	Earless	Earless
Position of Ear Leaves	Upright	Upright	Upright

Table 5. Leaf Morphological Characteristics of RGM1183 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Character	RGM1183	Clone 200	Clone 202	Clone 226
Curved Leaf Blade	1/3 - 1/2 Leaves	<1/3 Leaves	1/3 - 1/2 Leaves	<1/3 Leaves
Leaf Width	Narrow (<4cm)	Narrow (<4cm)	Narrow (<4cm)	Narrow (<4cm)
Leaf Color	Yellowish Green	Green	Yellowish Green	Green
Leaf Triangle Color	Green	Green	Green	Green
Leaf Ears	Earless	Earless	Earless	Earless
Position of Ear Leaves	Upright	Upright	Upright	Upright

Table 6. Leaf Morphological Characteristics of RGM186 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Character	RGM186	Clone 293	Clone 296
Curved Leaf Blade	1/3 - 1/2 Leaves	<1/3 Leaves	<1/3 Leaves
Leaf Width	Narrow (<4cm)	Narrow (<4cm)	Narrow (<4cm)
Leaf Color	Yellowish Green	Green	Green
Leaf Triangle Color	Green	Green	Green
Leaf Ears	Earless	Weak	Earless
Position of Ear Leaves	Upright	Upright	Upright

When observing the leaves, several characteristics can be observed, namely the curvature of the leaf, the width of the leaf, the color of the leaf, the color of the leaf triangle, the leaf ear, and the position of the leaf ear. Leaf blades of purple mutant clones of varieties RGM1183 and RGM186 are smaller than those of the original variety, i.e., less than 1/3 of the leaves. This means that it shows character differences from the original variety, indicating character diversity. According to Fathurrahman et al. (2024), colchicine induction can affect the characteristics of pulsative mutant leaves, one of which is leaf curl. However, the results of the study show that the leaf width of the original variety and the superior mutant clone is the same, with all leaves classified as narrow (<4 cm), indicating no significant variation in leaf width between the original variety and its superior mutant clones.

Leaf color characters differed between the original varieties and their superior mutant clones, except for GMP6, which was 100% yellowish-green (Table 3). Clones of RGM047 were 50% green and 50% yellowish-green. Clones of RGM1183 were 33% yellowish-green and 66% green. Clones of RGM186 were 100% green. These differences indicate the diversity between the original varieties and their superior mutant clones. Additionally, variations were observed in the triangular leaf characters: the GMP6 variety had purple triangular leaves, whereas clone 63 exhibited green triangular leaves. However, such diversity was not observed in RGM047, RGM1183, and RGM186 varieties or their mutant clones. According to Fathurrahman et al. (2024), in seeds treated

with colchicine, mitosis occurs in embryonic cells, followed by proplastid division and chromosome doubling. Colchicine induction can also inhibit chromosome separation during the anaphase stage, due to disruption in microtubule formation, which affects spindle fiber assembly (Mahfut et al., 2023c; 2023d). This process can lead to plants with higher chlorophyll levels, resulting in darker leaf coloration compared to the original variety.

In terms of ear leaf characteristics, differences were observed between the original varieties and superior mutant clones in GMP6 and RGM186, but not in RGM047 and RGM1183. The GMP6 variety possesses weak leaf ears, while clones 36 and 44 lack leaf ears entirely. Meanwhile, the RGM186 variety lacks leaf ears, whereas its superior mutant clone 293 exhibits weak leaf ears. These differences are attributed to the effects of colchicine, as colchicine induction is known to increase genetic diversity in plants and generate new trait variations (Yuniyati et al., 2022). However, colchicine treatment did not affect the position of the leaf ears, as no character differences in ear position were observed between the original varieties and their superior mutant clones.

The fourth parameter is the leaf midrib character. The leaf sheath morphological characteristics of GMP6, RGM047, RGM1183, and RGM186 and superior mutant clones at PT Gunung Madu Plantations are presented in Tables 7-10.

Table 7. Leaf Sheath Morphological Characteristics of GMP6 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Midrib Character	GMP6	Clone 36	Clone 44	Clone 63
Position of Dorsal Hair	Hairless	Hairless	Hairless	Hairless
Width of the Dorsal Hair Area	Hairless	Hairless	Hairless	Hairless
Distance to the Peak of the Dorsal Hair Area	Hairless	Hairless	Hairless	Hairless
Dorsal Hair Density	Hairless	Hairless	Hairless	Hairless
Ease of Leaf Detachment	Hard	Hard	Hard	Hard
Leaf Midrib Color	Purple	Purple	Purple	Purple

Table 8. Leaf Sheath Morphological Characteristics of RGM047 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Midrib Character	RGM047	Clone 160	Clone 162
Position of Dorsal Hair	Hairless	Hairless	Hairless
Width of the Dorsal Hair Area	Hairless	Hairless	Hairless
Distance to the Peak of the Dorsal Area	Hairless	Hairless	Hairless
Dorsal Hair Density	Hairless	Hairless	Hairless
Ease of Leaf Detachment	Currently	Currently	Currently
Leaf Midrib Color	Purple	Purple	Purple

Table 9. Leaf Sheath Morphological Characteristics of RGM1183 and Superior Mutant Clones at Gunung Madu Plantations

Leaf Midrib Character	RGM1183	Clone 200	Clone 202	Clone 226
Position of Dorsal Hair	Hairless	Hairless	Hairless	Hairless
Width of the Dorsal Hair Area	Hairless	Hairless	Hairless	Hairless
Distance to the Peak of the Dorsal Hair Area	Hairless	Hairless	Hairless	Hairless
Dorsal Hair Density	Hairless	Hairless	Hairless	Hairless
Ease of Leaf Detachment	Hard	Hard	Hard	Hard
Leaf Midrib Color	Green	Green	Purple	Purple

Table 10. Leaf Sheath Morphological Characteristics of RGM186 and Superior Mutant Clones at PT Gunung Madu Plantations

Leaf Midrib Character	RGM186	Clone 293	Clone 296
Position of Dorsal Hair	Lying down	Hairless	Hairless
Width of the Dorsal Hair Area	<1/4 width of leaf midrib	Hairless	Hairless
Distance from the top of the dorsal Hair Area	> 1 cm	Hairless	Hairless
Dorsal Hair Density	Sparse	Hairless	Hairless
Ease of Leaf Detachment	Easy	Easy	Hard
Leaf Midrib Color	Purple	Green	Green

When observing the leaf midrib, several characters were evaluated, including the position of the dorsal hairs, the width of the dorsal hair region, the distance to the apex of the dorsal hair region, dorsal hair density, the ease of leaf sheath detachment, and the color of the leaf midrib. Dorsal hair characters (position, width, distance, and density) are absent in all samples except for RGM186, which shows that its position is lying down, the width is <1/4 the width of the leaf midrib, the crest distance is >1 cm, and it has a sparse hair density. The loose (dry) nature of the leaf sheath is deep.

Variability in dorsal hair occurs only in the RGM186 variety. Dorsal hairs are needed to minimize stem borer attacks. According to Widiastuti et al. (2014), variety PS 882 had the

smallest stem borer attack rate among other varieties because it has broad dorsal hairs and an inclined position. For leaf sheath detachment, GMP6 and its clones, as well as RGM1183 and its clones, were difficult to detach, while variety RGM047 and its clones displayed moderate detachment. The variety RGM186 showed easy detachment, but one of its clones (296) was difficult to detach, indicating diversity induced by colchicine. Easily removed leaf sheaths are desirable for reducing labor costs in sugarcane cultivation (Pringgani et al., 2023; Mahfut et al., 2024c).

The leaf midrib color of the GMP6 and RGM047 varieties and their clones is purple, indicating no variation. Meanwhile, RGM1183 and RGM186 exhibited differences in leaf midrib color

compared to superior mutant clones. The RGM1183 had green midribs, while clones 202 and 226 were purple. Conversely, RGM186 displayed a purple midrib, while clones 293 and 296 were green. This suggests variation in leaf sheath color characters between the varieties RGM1183 and RGM186 and their superior mutant clones. The difference between green and purple in the leaf midrib is due to differences in the compounds they contain. This indicates that colchicine can change compounds in the midrib of plant leaves. According to Lv et al. (2021), plants turn purple because they contain anthocyanins, which can cause them to turn purple. This is also consistent with the opinion of Pringgani et al. (2023) that anthocyanins can cause purple, blue, red, and black colors in parts of rice plants.

The phenetic analysis was conducted to determine the relationship between the original variety and superior mutant clones. The similarity index based on morphological characters in original varieties and superior mutant clones at PT Gunung Madu Plantations is presented in Table 11, and the kinship dendrogram based on morphological characteristics of original varieties and superior mutant clones at PT Gunung Madu Plantations is shown in Figure 1.

Table 11. Similarity Index Based on Morphological Characters in Original Varieties and Superior Mutant Clones at PT Gunung Madu Plantations

	GMP6	CLONE36	CLONE44	CLONE63	RGM047	CLONE160	CLONE162	RGM1183	CLONE200	CLONE202	CLONE226	RGM186	CLONE293	CLONE296
GMP6	1.000													
CLONE36	0.929	1.000												
CLONE44	0.857	0.923	1.000											
CLONE63	0.929	0.857	0.786	1.000										
RGM047	1.000	0.929	0.857	0.929	1.000									
CLONE160	0.929	0.857	0.923	0.857	0.929	1.000								
CLONE162	0.857	0.786	0.846	0.786	0.857	0.923	1.000							
RGM1183	1.000	0.929	0.857	0.929	1.000	0.929	0.857	1.000						
CLONE200	0.714	0.643	0.692	0.643	0.714	0.769	0.833	0.714	1.000					
CLONE202	0.857	0.786	0.846	0.786	0.857	0.923	0.846	0.857	0.692	1.000				
CLONE226	0.714	0.643	0.692	0.643	0.714	0.769	0.833	0.714	1.000	0.692	1.000			
RGM186	1.000	0.929	0.857	0.929	1.000	0.929	0.857	1.000	0.714	0.857	0.714	1.000		
CLONE293	0.286	0.308	0.333	0.214	0.286	0.308	0.333	0.286	0.400	0.333	0.400	0.286	1.000	
CLONE296	0.286	0.214	0.231	0.214	0.286	0.308	0.333	0.286	0.400	0.333	0.400	0.286	0.600	1.000

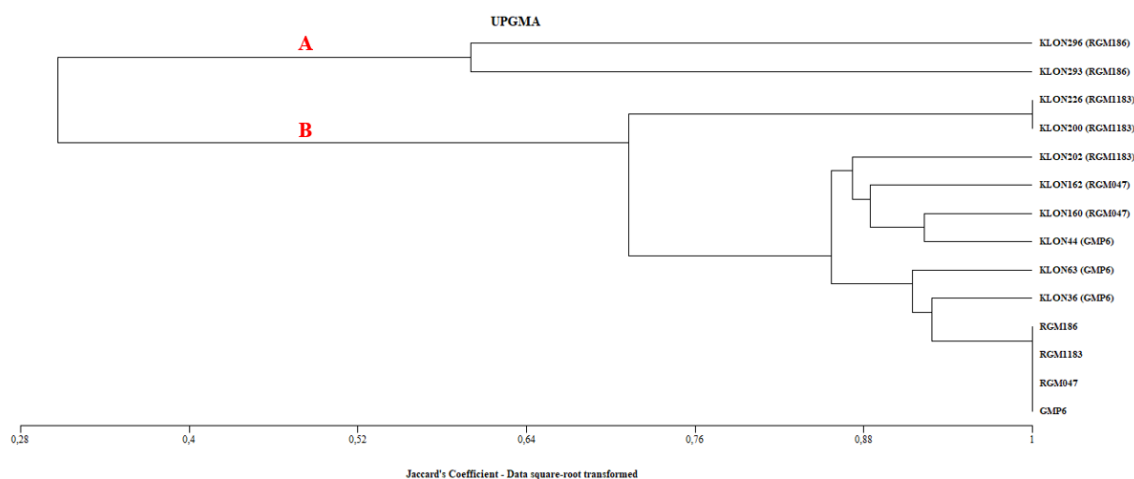


Figure 1. Kinship Dendrogram Based on Morphological Characteristics of Original Varieties and Superior Mutant Clones at PT Gunung Madu Plantations

Based on the dendrogram above, the samples are divided into two clusters, namely clusters A and B. Cluster A has a similarity index of 0.600 and consists of clones 293 and 296. Cluster A is divided into two subclusters: subcluster I (clone 293) and subcluster II (clone 296). Cluster B has a similarity index of 0.714 and consists of GMP6, RGM047, RGM1183, RGM186, clones 36, 63, 44, 160, 162, 202, and 226. Cluster B is divided into two subclusters, namely subcluster I, which consists of GMP6, RGM047, RGM1183, RGM186, clones 36, 63, 44, 160, 162, and 202, while subcluster II consists of clones 200 and 226.

Based on the similarity index (%), the closest qualitative agronomic characters are in group B, namely GMP6, RGM047,

RGM1183, RGM186, and clone 36, with a similarity index of 0.929. This is because the varieties and samples still have similarities in the qualitative agronomic characteristics observed in the field. Applying colchicine does not completely change the characteristics of the sample, but only improves it, so that the sample still has the same characteristics as the original variety. After knowing the relationship between the original varieties and mutant clones, a Principal Component Analysis (PCA) was conducted to identify the characteristics that influence the clustering. The results of the PCA based on morphological characters in original varieties and superior mutant clones at PT Gunung Madu Plantations are presented in Figure 2.

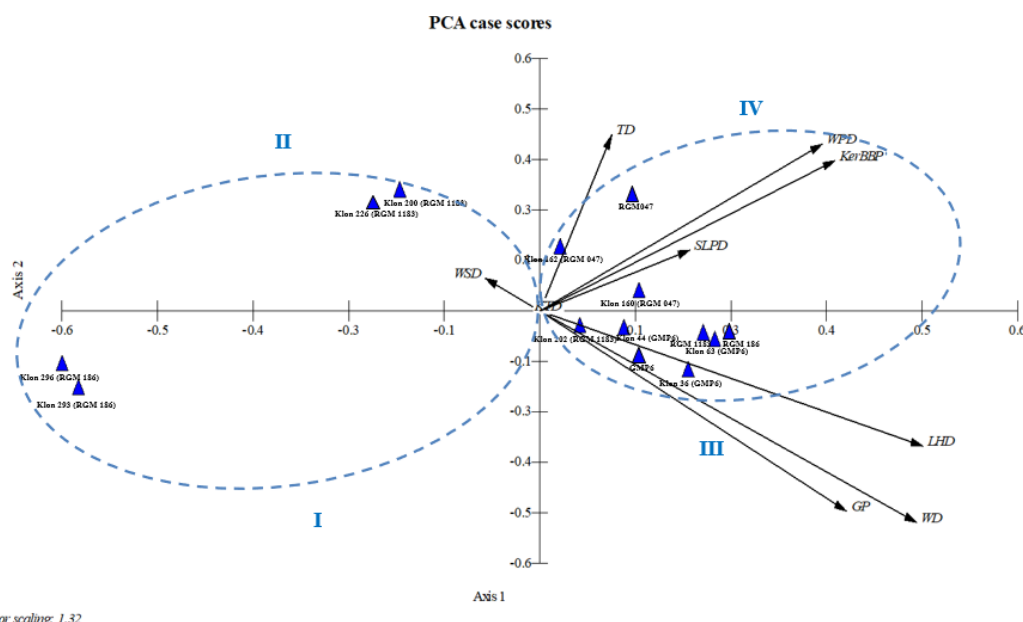


Figure 2. Principal Component Analysis (PCA) Based on Morphological Characters in Original Varieties and Superior Mutant Clones at PT Gunung Madu Plantations

The influential characters in grouping the original varieties and superior mutant clones at PT Gunung Madu Plantations can be seen in Table 12.

Table 12. Influential Characters in Grouping Original Varieties and Mutant Clones Superior at PT Gunung Madu Plantations

Code	Character	PC1	PC2	PC3	PC4
GP	<i>Grading</i> Germination	0.308	-0.382	0.568	0.517
GC	<i>Ground Coverage</i>	0.385	-0.257	-0.135	-0.299
LHD	Curved Leaf Blade	0.385	-0.257	-0.135	-0.299
LD	Leaf Width	0.000	0.000	0.000	0.000
WD	Leaf Color	0.379	-0.403	-0.054	-0.141
WSD	Leaf Triangle Color	-0.055	0.063	-0.217	-0.072
TD	Leaf Ears	0.072	0.336	0.715	-0.527
KTD	Position of Ear Leaves	0.000	0.000	0.000	0.000
KBBP	Position of Back Feathers	0.297	0.286	-0.077	-0.001
LDBBP	Width of the Dorsal Feather Area	0.297	0.286	-0.077	-0.001
JPDBBP	Distance to the Peak of the Dorsal Feather Area	0.297	0.286	-0.077	-0.001
KerBBP	Dorsal Feather Density	0.297	0.286	-0.077	-0.001
SLPD	Characteristics of Loose Leaf Sheath (Dry)	0.151	0.116	-0.232	0.181
WPD	Leaf Midrib Color	0.284	0.318	0.045	0.468
	<i>Eigen values</i>	1.170	0.313	0.188	0.150
	<i>Percentage</i>	58.972	15.787	9.483	7.574
	<i>Cum. Percentage</i>	58.972	74.759	84.242	91.816

Based on the results, PCA divided the samples into 4 distinct clusters based on their morphological characters. Cluster I consists of clones 293 and 296 that are grouped based on the position of the ear of the leaf (KTD). Cluster II consists of clones 200 and 226, which are grouped based on similarity of leaf ear position (KTD) and leaf triangle color (WSD). Cluster III consists of clones 202, 44, GMP6, RGM1183, RGM186, clones 63 and 36, which are grouped based on leaf ear position similarity (KTD), germination grading (GP), leaf color (WD), and leaf blade curvature (LDH). Cluster IV consists of RGM047, clones 160 and 162, which are grouped based on the similarity of leaf ear position (KTD), leaf ear (TD), leaf midrib color (WPD), dorsal hair density (KerBBP), and leaf sheath laxity (SLPD).

Based on the PCA results, there is grouping in the sample, which is divided into 4 clusters that are influenced by the characters in the sample and can be seen in Table 12, which shows the influence of each character, which can be seen from the large or small eigenvalues. Based on this, it can be seen that the characters that influence all samples are those that have a value of 0.000, namely Leaf Width (LD) and Position of Ear Leaves (KTD). According to Mahfut et al. (2024a), apart from the cluster grouping pattern (I, II, III, and IV), the characters in the superior mutant GMP3 also play a role in separating samples between cluster analysis and principal component similarity analysis in which there are similarities.

In the PCA above, the eigenvalues show the percentage contribution of each group. Axis I accounts for 58.97% with a value of 1.170, Axis II is 15.78% with a value of 0.313, Axis III is 9.48% with a value of 0.118, and Axis IV is 7.57% with a value of 0.150. This eigenvalue shows the influence of each character present.

4. Conclusion

The grouping of samples is based on similar characteristics. These similarities strengthen the kinship relationships among the samples, which enhances the accuracy of classification. Colchicine induction consistently improves various morphological traits across all samples, including increased leaf width and position of ear leaves, which contribute to enhanced photosynthetic efficiency. The findings obtained highlight the effectiveness of colchicine as a mutagenic agent for inducing genetic variation in sugarcane. The research demonstrates that colchicine treatment can significantly improve the agronomic traits of sugarcane, offering valuable insights for future breeding programs aimed at enhancing productivity. Future studies should explore the long-term effects of colchicine on sugarcane yield and investigate the molecular mechanisms underlying these agronomic improvements.

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7. References

- Anbiya L, Mahfut, Wahyuningsih S, Suratman (2024). Optimum temperature for detection of TrAP and Rep genes using SPG1/SPG2 primer design on red chili (*Capsicum annuum* L.). The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050034. <https://doi.org/10.1063/5.0208181>.
- Aritonang LL, Mahfut, Ahyar AN (2024). Acclimatization of five commercial varieties of sugarcane (*Saccharum officinarum* L.) at PT. GunungMadu Plantations, Lampung. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050004. <https://doi.org/10.1063/5.0208181>.
- Asadudin D, Mulya IP, Nugraha MA, Hasanudin SNA, Mahfut, Susiyanti E, Ahyar AN (2024). Anatomic characterization of 24 mutant varieties of GMP6 in PT. GunungMadu Plantations. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050039. <https://doi.org/10.1063/5.0208463>.
- Fathurrahman F, Ulpah S, Sodik Nam, Mahadi I (2024). The effect of colchicine treatment on phenotype and genotype characteristics of Detam-2 variety of soybean *Glycine max*. *Biodiversitas.* 25(3): 1230-1238. <https://doi.org/10.13057/biodiv/d250339>.
- Food and Agriculture Organization (2020). Current world fertilizer trends and outlook for 2018. Food and Agriculture Organization of the United Nations. Rome.
- Lv Z, Zhu F, Jin D, Wu Y, Wang S (2021). Seed germination and seedling growth of *Dendrocalamus brandisii* in vitro, and the inhibitory mechanism of colchicine. *Front Plant Sci.* 12: 784581. <https://doi.org/10.3389/fpls.2021.784581>.
- Mahfut (2020). Identification and detection *Odontoglossum ringspot virus* on native orchids collection of nurseries in Java, Indonesia. *J. Phys. Conf. Ser.* 1641(012075): 1-6. <https://doi.org/10.1088/1742-6596/1641/1/012075>.
- Mahfut (2021). Morphological identification of mycorrhizal fungi isolated from native orchid in Indonesia. *Jordan J. Biol. Sci.* 14(5): 1031-1034. <https://doi.org/10.54319/jjbs/140523>.
- Mahfut, Ananda M, Susiyanti E, Agustina R (2025a). Screening of sugarcane commercial GMP varieties tolerant to drought stress based on molecular detection of P5CS gene. *J. Multidiscip. Appl. Nat. Sci.* 5(1): 119–129. <https://doi.org/10.47352/jmans.2774-3047.233>.
- Mahfut, Angeiny A, Wahyuningsih S, Handayani TT, Sukimin (2020a). Identification of disease and efforts to protect native orchid plants against bacterial infection in Liwa Botanical Garden. *J. Phys. Conf. Ser.* 1641 (012098): 1-8. <https://doi.org/10.1088/1742-6596/1641/1/012098>.
- Mahfut, Handayani TT, Wahyuningsih S, Sukimin (2021a). Identification of *Dendrobium* (Orchidaceae) in Liwa Botanical Garden based on leaf morphological characters. *J. Trop. Biodivers.* 6(1): 1-6. <https://doi.org/10.22146/jtbb.59423>.
- Mahfut, Hidayat MM, Arifannisa SJ (2023a). Study of orchid resistance induction using *Rhizoctonia* against ORSV infection based on anatomical characters of roots and leaves. *Asian J. Plant Sci.* 22(2): 239-249. <https://doi.org/10.3923/ajps.2023.239.249>.
- Mahfut, Indrianto A, Somowiyarjo S, Daryono BS (2020b). Molecular phylogeny of orchids mycorrhiza isolated from native tropical orchids in Indonesia. *Malays. J. Microbiol.* 16(1): 68-72. <https://doi.org/10.21161/mjm.190425>.
- Mahfut, Kendari P, Bangsawan R, Susiyanti E (2024a). Agronomic characteristics of sugarcane cultivar GMP3 mutants induced through colchicine. *SABRAO J. Breed. Genet.* 56(3): 1083-1094. <https://doi.org/10.54910/sabrao2024.56.3.16>.
- Mahfut, Kendari P, Susiyanti E, Ahyar AN, Bangsawan R (2024b). Molecular characterization of the sugarcane cultivar GMP3 mutants induced through colchicine using PCR-RAPD markers. *SABRAO J. Breed. Genet.* 56(3): 940-950. <https://doi.org/10.54910/sabrao2024.56.3.4>.
- Mahfut, Kendari P, Syarif A, Wahyuningsih S, Susiyanti E (2023b). Stomata characters of sugarcane (*Saccharum officinarum* L.) mutants of GMP3 variety at PT Gunung Madu Plantations, Lampung, Indonesia. *J. Trop. Biodivers. Biotechnol.* 8(3): 1-12. <https://doi.org/10.22146/jtbb.79860>.
- Mahfut, Minarni IY, Wahyuningsih S, Handayani TT (2021b). Physiological analysis of orchid chlorophyll against *Odontoglossum ringspot virus* infection. *Jordan J. Biol. Sci.* 14(5): 1025-1029. <https://doi.org/10.54319/jjbs/140522>.
- Mahfut, Riniarti M, Kurniawaty E (2025b). Morphology and phytochemical characterization of native *Dendrobium* used as a herbal medicine in Lampung, Indonesia. *SABRAO J. Breed. Genet.* 57(2):587-598. <https://doi.org/10.54910/sabrao2025.57.2.17>.
- Mahfut, Sari HE, Kendari P (2024c). Habitat identification and phenetic analysis of bryophyta in Lampung, Indonesia. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050014. <https://doi.org/10.1063/5.0208187>.
- Mahfut, Sari M, Akin HM, Wahyuningsih S, Irawan B (2025c). Molecular characterization and effectiveness cross-protection of weak strains against super-infection malignant strains

- Cucumber mosaic virus* (CMV) on cayenne pepper (*Capsicum annuum* L.) in Lampung, Indonesia. *J. Trop. Plant Pests Dis.* 25(1): 63-75. <https://doi.org/10.23960/jhptt.12563-75>.
- Mahfut, Setiawan A, Sari M, Sijabat VE, Siregar VAP, Ahmad Z (2023c). Molecular characterization of mycorrhiza and its potential as biocontrol. *SABRAO J. Breed. Genet.* 55(6): 2092-2104. <https://doi.org/10.54910/sabrao2023.55.6.21>.
- Mahfut, Syahira H, Wahyuningsih S, Handayani TT, Sukimin (2020c). Identification of virus infection on native orchids in Liwa Botanical Garden. *J. Phys. Conf. Ser.* 1751(012063): 1-7. <https://doi.org/10.1088/1742-6596/1751/1/012063>.
- Mahfut, Tolangara A, Ahmad H, Rasyid R, Ahmad Z (2023d). Variation of resistance and physiological response of orchid from induction *Trichoderma* to infection *Odontoglossum ringspot virus* (ORSV). *Asian J. Plant Sci.* 22(3): 496-505. <https://doi.org/10.3923/ajps.2023.496.505>.
- Nuraini S, Mahfut, Bangsawan R (2024). Germination of bud chips 5 varieties of commercial sugarcane (*Saccharum officinarum* L.) at PT. GunungMadu Plantation. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050037. <https://doi.org/10.1063/5.0208193>.
- Pringgani AM, Khonghintaiong J, Gonkhamdee S, Songsri P, Jongrunklang N (2023). Ground covering characteristics of sugarcanes using high-angle images and their relationship with growth destructive sampling. *Asian J. Plant Sci.* 22: 434-443. <https://doi.org/10.3923/ajps.2023.434.443>.
- Putera AY, Mawardi I, Ananda M, Sephanti DI, Rajagukguk OPU, Pubianty DP, Mahfut, Ahyar AN, Bangsawan R, Susiyanti E (2024). Anatomic stomata characteristics of 24 mutants of RGM 01-1869 varieties in PT. Gunung Madu Plantations. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050038. <https://doi.org/10.1063/5.0208535>.
- Putra AS, Syarif A, Mahfut, Sulistiyanti SR (2024a). One decade research in the field of artificial intelligence for Indonesian orchid: a bibliometric analysis. *Afr. J. Biol. Sci.* 6(5): 10848-10860. <https://doi.org/10.48047/AFJBS.6.5.2024.10848-10860>.
- Putra AS, Syarif A, Mahfut, Sulistiyanti SR, Hasibuan M (2024b). Performance evaluation of fuzzy logic system for *Dendrobium* identification based on leaf morphology. *J. Appl. Data Sci.* 5(4): 1520-1529. <https://doi.org/10.47738/jads.v5i4.224>.
- Sadiq NM, Robinson KJ, Terrell JM (2025). Colchicine. StatPearls Publishing: Treasure Island (FL).
- Sari HE, Mahfut, Widyawan A, Irawan B, Wahyuningsih S (2024). Analysis of total DNA quality and quantity in commercial varieties of sugarcane (*Saccharum officinarum* L.) at PT GunungMadu Plantations. *Afr. J. Biol. Sci.* 6(13): 6095-6101. <https://doi.org/10.48047/AFJBS.6.13.2024.6095-6101>.
- Sari HE, Mahfut, Widyawan A, Wahyuningsih S, Irawan B (2025). Sugarcane commercial cultivars with drought stress tolerance on in vitro and greenhouse scales. *SABRAO J. Breed. Genet.* 57(1): 137-148. <https://doi.org/10.54910/sabrao2025.57.1.14>.
- Septiana AZ, Mahfut, Handayani TT, Suratman (2024). Survey of Tomato yellow leaf curl virus (TYLCV) infection on *Solanum lycopersicum* L. in Lampung, Indonesia. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050027. <https://doi.org/10.1063/5.0208190>.
- Simamora S, Mahfut, Bangsawan R (2024). Germination test of commercial variety of sugarcane (*Saccharum officinarum* L.) in the end planting month using bud chips at PT GunungMadu Plantations. The 3rd Universitas Lampung International Conference on Science, Technology and Environment (ULICoSTE) 2022. *AIP Conf. Proc.* 2970: 050029. <https://doi.org/10.1063/5.0208191>.
- Sukmawati S, Badaruddin I, Dewi NK, Situmorang N, Mahfut, Mustapa E (2021). Analysis of organoleptic and coliform value in fresh mackerel (*Rastrelliger* sp.) fish in TPI Sorong city. *J. Phys. Conf. Ser.* 1764(012035): 1-6. <https://doi.org/10.1088/1742-6596/1764/1/012035>.
- Yuniyati N, Trikoesoeningtyas, Hartati RS, Suhesti S (2022). Colchicine induces morphological variability in sugarcane. *IOP Conf. Ser.: Earth Environ. Sci.* 974(012043). <https://doi.org/10.1088/1755-1315/974/1/012043>.
- Windiyani IP, Mahfut, Purnomo, Daryono BS (2022). Morphological variations of superior sugarcane cultivars (*Saccharum officinarum*) from Lampung, Indonesia. *Biodiversitas.* 23(8): 4109-4116. <https://doi.org/10.13057/biodiv/d230831>.