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Morphology, antibacterial, and molecular analysis using the RAPD method on bamboo in Bengkulu, Indonesia

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Abstract The Bengkulu community uses bamboo shoots for fermented food lemea, a local fermented food. Conservation is crucial to prevent extinction. A study explored bamboo's shape, antibacterial qualities, and genetic features. Bamboo samples were collected and morphological properties and antibacterial activity were identified. The study found differences in morphological, antibacterial, and molecular similarity coefficients. The similarity coefficient based on morphological characters was 59%, divided into two clusters. Based on the results of molecular analysis, the similarity coefficient was 79%, divided into three clusters. There were differences in morphological, antibacterial and molecular similarity coefficients.

Keywords: Antibacterial, Bamboo, Diversity, Molecular, Morphology, RAPD

Introduction

Bamboo is included in the angiosperm group of the monocot order, with the appearance of tree-like plants and grasses that from the subfamily Bambusoideae of the grass family Poaceae. There are more than 116 genera and 1,439 accepted species of bamboo, of which Indonesia has 26 genera with 174 bamboo species (Widjaja *et al*., 2020). Out of 4 generas and 13 species were found in Bengkulu Tengah; *Bambusa (B.) glaucescens, B. multiplex*, *B. vulgaris var. vulgaris*, *B. vulgaris var. striata*, *Dendrocalamus (D.) asper*, *Gigantochloa (G.) apus*, *G. hasskarliana, G. pseudoarundinaceae*, G. *serik, G. scortechinii, G. robusta, Schyzotyum (S.) brachycladum, and S. lima* (Hastuti *et al.,* 2018; Yani and Anggraini, 2018).

Using bamboo for food is one of its advantages. Some species are usually used for edible purposes including *B. vulgaris*, *D. asper*, *G. apus*, *G.*

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atroviolacea, *G. atter*, and *G. pseudoarundinacea* (Kumalasari *et al*., 2019), and the parts usually consumed are bamboo shoots with one of the processed products, namely lemea (Peronika *et al*., 2022). While other plants take a long time to mature, bamboo only takes 3-6 years, at which time bamboo has matured and is usually used for building construction (Feng *et al*., 2023; Yadav and Mathur, 2021).

The morphology of the characters varies depending on the age and height of the culm; among them are internode length, outer internode diameter, and culm wall thickness (Widjaja *et al*., 2005; Gu *et al*., 2019). Based on the morphological identification key, there are 39 species from 12 bamboo genera in Sulawesi (Ervianti *et al*., 2019). Bamboo from the Indrokilo botanical garden, Central Java, based on its morphological characters, were found to have 5 clusters with a similarity coefficient of 70% (Wahidah *et al.,* 2021). Weh Island, Aceh, has 8 species with 4 clusters and a similarity coefficient of 60% (Ritonga *et al*., 2023). The morphological characteristics of bamboo plants consist of quantitative and qualitative characters (Widjaja *et al*., 2020). Differences in morphological characters can be employed as a basis for exploring the potential of bamboo.

Genetic diversity represents the heritable variation both within and among populations of organisms as well as shows important implications for the evolution and conservation of species (Ellegren and Galtier, 2016). The approaches through molecular markers were known to have a higher level of accuracy compared to the morphological approach in the identification and grouping of bamboo cultivars (Ramanatha Rao and Hodgkin, 2002; de Jesus *et al*., 2013; Hapsari *et al*., 2015; Maftuchah *et al.*, 2021). One of the molecular markers applied in research on genetic diversity in plants was Random Amplified Polymorphic DNA (RAPD) (Annisa *et al*., 2019); Probojati *et al.,* 2019). The RAPD were the first PCR-based molecular markers used in genetic variation analysis. The advantages of using RAPD include low cost, fast pace, producing many polymorphic, and being sensitive in determining the genetic relationships between species or individuals (Amom *et al*., 2020).

Unfortunately, there are still very few cultivation efforts from the community to preserve bamboo, and they are yet harvested from nature freely without re-establishing the stands by selective harvesting (Yani and Anggraini, 2018). Relationship analysis in many categories can be seen using the PCA and clustering methods (Reddy *et al*., 2020; Oggier and Datta, 2023). Studying the genetic diversity of bamboo plants in Bengkulu using RAPD molecular markers is very important for breeding. The study aimed to determine the genetic diversity of bamboo in Bengkulu based on morphologicaland molecular characteristics

using RAPD markers for the conservation of bamboo plants as raw materials for fermented food.

Materials and methods

Plant materials

This study examined the morphological characteristics of 13 bamboo species taken from four regions in Bengkulu Province based on both quantitative and qualitative characteristics. This study also examined the diversity of 13 bamboo species with the RAPD molecular marker. Geographical distribution of the sampled districts is visually depicted (Figure 1), as illustrated in the regional map.

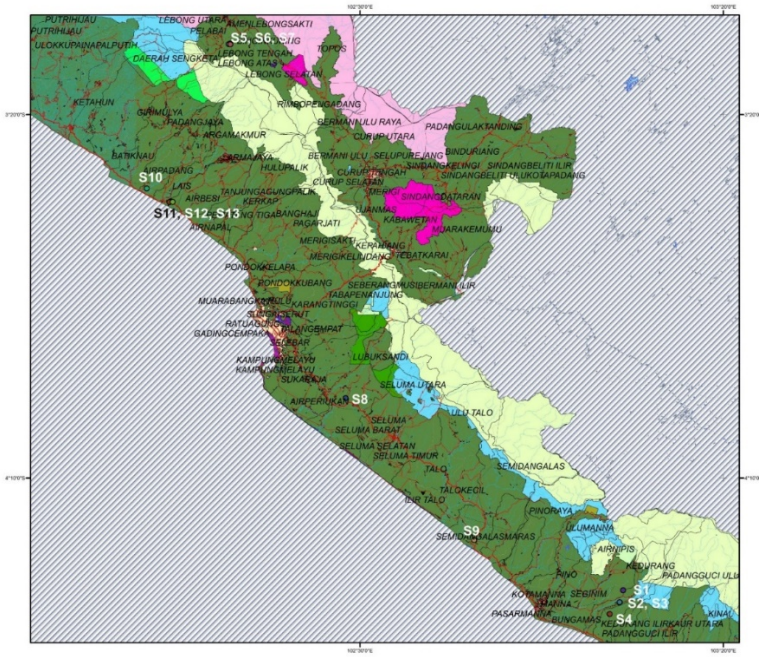


Figure 1. Map of bamboo sampling areas in Bengkulu

Morphological analysis

Bamboo morphology analysis was carried out quantitatively (using measurements) on stems and qualitatively (doing visual observation) on stems, leaves, rhizomes, branches, shoots, and feathers. The observed bamboo sample criteria were: bamboo grown in Bengkulu in four districts (South Bengkulu,

Lebong, Seluma, and North Bengkulu) from which the bamboo shoots are consumed by the local community. The bamboo stems criteria are standing upright, mature, having complete leaves to the tip of the stem, hard, mature bamboo stems falling off easily, and aged ± 3-6 years. Bamboo culms are measured in three parts: at the base (5th node), middle (15th node), and tip (25th node) (Yadav and Mathur, 2021). The tools used in morphological analysis were a knife, meter, and sigmat.

Qualitative observations included stem (distance between stems, presence of segments, surface of the segments, surface of the stem, shape of the bamboo segments, and contents of the bamboo segments); leaves (characteristics, color, shape, and size of the leaves); rhizomes (length and diameter rhizomes, rhizome characteristics, and root location); branches (branch position, number of branches, location of branch emergence, branch posture, and branch modifications); shoots and hairs. The bamboo leaves observed in this research were bamboo leaves on the first branch that were fresh and undamaged; the color had not turned yellow due to age; and the branches were seen at the bamboo stem segment containing the first branch (Damayanto and Widjaja, 2017).The outcomes of observations were documented and shared in Excel as codes or numbers for data analysis.

Antibacterial activity analysis: Stages of making simplicia: Bamboo shoots were thinly sliced and dried using a cabinet drying machine, then ground to obtain simplicia powder.

Extraction Stage: Maceration was carried out once by means of 250 g of simplicia powder soaked in 2500 mL of 96% ethanol (1:10 ratio) for 5 days with occasional stirring, then filtered using filter paper to produce filtrate. The simplicia filtrate was thickened using a ratory evaporator at 40 °C so that a black-brown viscous extract with a distinctive aroma was obtained.

Antibacterial activity test stage: the antibacterial activity test used 13 sample extracts, a positive control was tetracycline, and a negative control used distilled water, which were tested on pathogenic bacteria, namely *Staphylococcus aureus*. Each treatment was replicated in three petri disk The pathogenic bacteria were a pure culture of *Staphylococcus aureus*, which was obtained from the University of Indonesia, with a gram-positive form, a coccus, a stapylo arrangement, non-spore-forming, and non-motile, which were later added to TSA media for antibacterial testing.

Antibacterial test: 1 gram of sample was extracted on paper discs and until 1 g of sample was used up; the paper discs were left for 1 min and placed on the agar media, which contained pathogenic bacteria; The positive control used a tertasicline solution with a concentration of 30 g/ml, and the negative control used a sterile distilled water solution. The discs were soaked for 1 min in the

positive and negative control solutions; the cup was filled with a mixer three times for each sample; and the cup was incubated at 37 °C for 1x24 h. Antibacterial activity is stated as positive if an inhibition zone is formed in the form of a clear zone around the disc paper. The diameter of the inhibition zone was measured using a caliper.

Molecular analysis

The stages of molecular analysis are DNA extraction, amplification, and electrophoresis. DNA was extracted from 13 bamboo samples using a modified CTAB (Cetyl Trimethyl Ammonium Bromide) method (Doyle, JJ; Doyle, 1990) A total of 0.1 g of bamboo leaf sample was crushed using a mortar into fine powder and added with 10 ml of liquid nitrogen; 500 µl of CTAB buffer solution was added to the samples, mixed and incubated at 65 °C for 60 minutes, and stirred every 10 minutes. The mixed sample was incubated at room temperature for 2 minutes, 500 µL CIA (Cloroform isoamyl alcohol, 24: 1) was added to the sample then was centrifuged at 12,000 rpm for 15 minutes. 300 µL of supernatant was taken and transferred to a new 2 mL tube; sodium acetate was added at a ratio of 1:10 of the volume and isopropanol was added using 2:3 of the supernatant volume; than incubated at -20 ^{0}C for 24 h than room temperature for 1 min. The supernatant was centrifuged again at 12,000 rpm for 10 minutes; than of the DNA pellets were obtained than cleaned with 500 µL 70% ethanol and centrifuged at 8,000 rpm for 5 minutes; the supernatant was carefully removed and the the DNA pellet was air dried for 20 minutes. 50 µL of nuclease-free water were finally added to the dry DNA pellet.

Testing resulted in the selection of 81 primers for amplification, leaving 8 primers usable: OPA 1, OPA 8, OPA 16, OPC 11, OPD 20, OPM 4, OPM 12, and OPM 14. Amplification reaction was performed by mixing 0.1 µL DNA template, 1 µLforward and reverse primers, 9.5 µL nuclease-free water, and 1 12.5 µL green Taq DNA polymerase. 0.5 µL PCR products were analyzed using gel electrophoresis by 1% agarose 1x TAE buffer using10000 bp DNA ladder. Electrophoresis was carried out using Mupid Exu Electrophoresis for 50 minutes with a tension of 50Vv, then the agarose gel was soaked with diamond nucleic acid liquid for 15 minutes and visualized using an ultra-slim LED illuminator with sensitivity down to 0.5 ng, uniformity <10% CV, illumination wavelength ~470 nm.

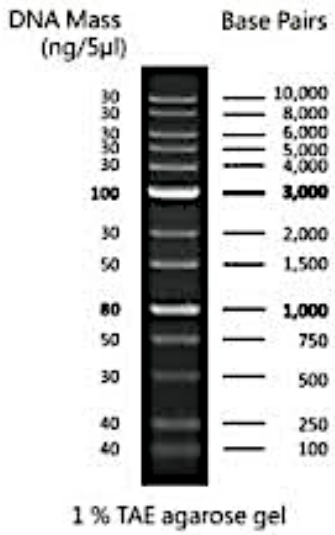


Figure 2. Marker of DNA ladder

Data analysis

The morphological characterisation data, both quantitative and qualitative, were provided in a descriptive manner. Using information from qualitative morphological characterisation, a morphological key was created. Based on morphological traits, antibacterial and molekuler the data were then examined for bamboo diversity using PCA with XL-Stat and clustering using NTSYS-pc 2.0 UPGMA.

Results

Sampling was carried with measurements of altitude, relative humidity, and scientific names based on local names (Table 1).

Table 1. Bamboo species in Bengkulu, Indonesia

Sample code	Local name	Scientific name	Status	Village, Regency	Altitude	Relative Humidity
S1	Buluh aur kuning	*Bambusa vulgaris Schrad. ex* J.C.Wendl	China (Yunnan) to Indo-China	Palak Siring, Bengkulu Selatan	167	80%
S2	Buluh mayan	*Gigantochloa robusta* Kurz	Native	Tanjung Alam, Bengkulu Selatan	143	80%

Sample code	Local name	Scientific name	Status	Village, Regency	Altitude	Relative Humidity
S3	Buluh Dabuk	*Gigantochloa atter* (Hassk.) Kurz ex Munro	Native	Tanjung Alam, Bengkulu Selatan	144	80%
S4	Buluh Betung	*Dendrocalamus asper* (Schult.& Schult.f) Backer	Native	Durian Sebatang, Bengkulu Selatan	230	80%
S5	Bambu Kapea	*Schizostachyum brachycladum* (Kurz ex Munro) Kurz	Indo-China to W. & Central Malesia	Pagar Agung, Lebong	1100	65%
S6	Bambu Seik	*Gigantochloa serik* Widjaja	Native	Pagar Agung, Lebong	1100	65%
S7	Bambu Dabuk	*Gigantochloa atter* (Hassk.) Kurz ex Munro	Native	Pagar Agung, Lebong	1100	65%
S8	Bambu Mayan	*Gigantochloa robusta* Kurz	Native	Gunung Agung, Seluma	20	85%
S9	Bambu Kapal	*Schizostachyum brachycladum* (Kurz ex Munro) Kurz	Native	Kembang Mumpo,Seluma	32	85%
S10	Bambu Mayan	*Gigantochloa robusta* Kurz	Native	Dusun Raja, Bengkulu Utara	6	85%
S11	Bambu Serik	*Gigantochloa serik* Widjaja	Native	PAL 30, Bengkulu Utara	15	95%
S12	Bambu Betung	*Dendrocalamus asper* (Schult.& Schult.f) Backer	Native	PAL 30, Bengkulu Utara	15	95%
S13	Bambu Aur	*Bambusa vulgaris Schrad. ex* J.C.Wendl	Native	PAL 30, Bengkulu Utara	15	95%

Morphological character analysis

It provided information on the morphological characteristics of bamboo, including qualitative and quantitative characters (Figure 3). The analysis of the morphological characteristics of the 13 bamboos found that the S11 (*Gigantochloa serik,* Widjaja) bamboo culms had the longest internode, namely 34.0 ± 8.3 cm; while the S2 (*Gigantochloa robusta* Kurz) bamboo had the shortest internode, which was 19.5 ± 1.0 cm (Table 1). S12 (*Dendrocalamus asper* (Schult.& Schult.f) Backer) bamboo had the highest internode thickness of 2.97 cm; while S9 bamboo had the smallest internode thickness of 0.37 cm; and the highest bamboo internode diameter was 28.23 cm in S2 and 9.33 cm in S5 (*Schizostachyum brachycladum* Kurz ex Munro).

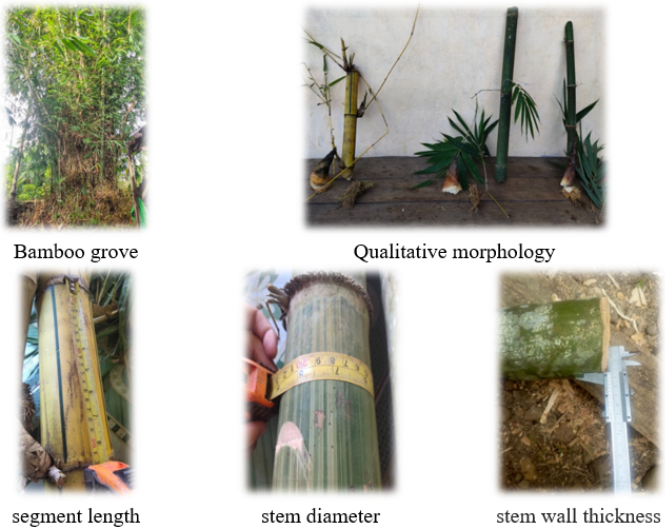


Figure 3. Morphological analysis of bamboo

Table 2. Morphological characters of 13 species of bamboo in Bengkulu

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
S1 *Bambusa vulgaris var. striata* (Lodd.Ex Lindl.) Gamble	38.8 ± 0.6	20.9 ± .7	1.3 ± 0.2	Stems formed dense clumps; nodes were solitary, not smooth, surface of internodes rough below; dusty cavities; hanging leaves- green on both sides long; rhizome short and thick; branches all over the stem, more than three

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
				branches, and one of the branches was larger; branches found above the nodes, horizontal and thornless, shoots covered with feathers, and the color of the miang was black and even.
S2 *Bambusa vulgaris Schrad.* ex J.C.Wendl	19.5 ± 1.0	28.2 ± 8.2	2,6 ± 0.2	Stems formed dense clumps with solitary nodes, a rough surface at the bottom, dusty cavities; erect leaves, green on both sides, wide; the rhizome short and thick; branches all over the stem, more than three branches, and one of the branches is larger, above nodes, angled upwards, had spines, shoots were covered with feathers, the color of the miang was black and even.
S3 *Gigantochloa atter* (Hassk.) Kurz ex Munro	25.7 ± 0.2	17.0 ± 4.6	1.4 ± 0.1	Stems formed dense clumps, nodes were tight, the surface of the segments was smooth as it went upwards, the cavity was dusty, the leaves were hanging, green on both sides, wide; rhizomes were short and thick;branches at the top of the stem, It has more than three branches, one of the branches is larger, the branches are above the nodes, the shoots are covered with wax, the shoots are slightly hairy with brown hairs, shoots were covered with hair and wax; the color of the miang was brown and uneven.
S4 *Dendrocalamus asper* (Schult.& Schult.f) Backer	22.9 ± 0.4	19.2 ± 3.6	1.7 ± 0.1	Stems formed dense clumps, smooth surface of internodes, dusty cavities; hanging leaves, lighter on one side, long; rhizome wasshort and

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
				thick, solitary node, creeping along the surface; branches at the top of the stem, more than three branches and a larger one of the branches, branch on the node line, angled upwards, had no thorns; shoots were covered with hairs; the color of the miang was black, smooth, and just a little hair.
S5 *Schizostachyum brachycladum* (Kurz ex Munro)	32.7 ± 1.7	9.3 ± 1.8	0.4 ± 0.1	Stems formed clumps, nodes were solitary, the surface of the segments was smooth as it went upwards, the cavity was dusty; the leaves were hanging, green on both sides, wide; the rhizomes were long and thin; branches were at the top of the stem, more than three branches, all in the same size: branches were above the node, angled upwards, had no spines, shoots were covered with feathers; the color of the miang were black, smooth, and in small number
S6 *Gigantochloa serik* Widjaja	25.2 ± 6.7	16.3 ± 3.3	1.5 ± 0.2	Stems formed dense clumps, nodes were tight, the surface of the internodes was rough below, the cavity was dusty, the leaves were hanging, brighter on one side, long;the rhizomes were long and thin; branches at the top of the stem, more than three branches, and one of the branches was larger, branches were above the node, angled upwards, had no thorns; purplish green shoots covered with hairs, and the color of the miang was brown and uneven.

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
S7 *Bambusa vulgaris* Schrad. ex J.C.Wendl	31.4 ± 5.8	24.4 ± 8.0	1.5 ± 0.1	Stems formed dense clumps with solitary nodes, a rough surface at the bottom, dusty cavities; erect leaves, green on both sides, wide; the rhizome was short and thick; branches all over the stem, more than three branches, and one of the branches was larger, above nodes, angled upwards, had spines, shoots were covered with feathers, the color of the miang was black and even.
S8 *Bambusa vulgaris* Schrad. ex J.C.Wendl	29.8 ± 6.9	16.5 ± 1.5	1.2 ± 0.4	Stems formed dense clumps with solitary nodes, a rough surface at the bottom, dusty cavities; erect leaves, green on both sides, wide; the rhizome was short and thick; branches all over the stem, more than three branches, and one of the branches was larger, above nodes, angled upwards, had spines, shoots were covered with feathers, the color of the miang was blackand even.
S9 *Schizostachyum brachycladum* (Kurz ex Munro) Kurz	31.3 ± 1.8	12.0 ± 1.6	0.4 ± 0.0	Stems formed dense clumps, nodes are solitary, the surface of the segments is smooth as it goes upwards, the cavity is dusty, the leaves were hanging, green on both sides, wide; the rhizomes were long and thin, branches at the top of the stem, more than three branches and in the same size, branches above the node, angled upwards, had no spines, shoots were covered with feathers, the color of the miang was black, smooth, and in small number

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
S10 *Bambusa vulgaris* Schrad. ex J.C.Wendl	30.9 ± 7.0	22.4 ± 8.5	1.2 ± 0.5	Stems formed dense clumps with solitary nodes, a rough surface at the bottom, dusty cavities, erected leaves, green on both sides, wide, the rhizome was short and thick, branches all over the stem, more than three branches, and one of the branches was larger, above nodes, angled upwards, had spines, shoots were covered with feathers, the color of the miang was black and even.
S11 *Gigantochloa serik* (Widjaja)	34.0 ± 8.3	16.5 ± 3.8	1.1 ± 0,1	Stems formed dense clumps, nodes were tight, the surface of the internodes was rough below, the cavity was dusty, the leaves were hanging, brighter on one side, long; the rhizomes were long and thin, branches at the top of the stem, more than three branches, and one of the branches was larger, branches above the node, angled upwards, had no thorns, purplish green shoots covered with hairs, and the color of the miang was brown and uneven.
S12 *Dendrocalamus asper* (Schult.& Schult.f) Backer	31.4 ± 6.7	26.2 ± 8.5	3.0 ± 0.3	Stems formed dense clumps, smooth surface of internodes, dusty cavities, hanging leaves, lighter on one side, long; rhizome was short and thick, solitary node, creeps along the surface, branches at the top of the stem, more than three branches and a larger one of the branches, branch on the node line, angled upwards, has no thorns, shoots were covered with

Sample code	Average segment length ± SE (cm)	Average stem diameter ± SE (cm)	Average stem wall thickness ± SE (cm)	Qualitative Analysis
				hairs, the color of the miang was black, smooth, and just a little hair.
S13 *Gigantochloa atter* (Hassk.) Kurz ex Munr	24.6 ± 5.2	18.5 ± 5.3	0.4 ± 0.0	Stems formed dense clumps, nodes were tight, the surface of the segments was smooth as it went upwards, the cavity was dusty, the leaves were hanging, green on both sides, wide; rhizomes were short and thick, branches at the top of the stem, more than three branchesand one branch is bigger than the other branches. . One branch, branch, branch above the node, horizontal, had thorns; shoots were covered with hair and wax; the color of the miang was brown and uneven.

Antibacterial character analysis

Bamboo shoots from the 13 bamboo samples were extracted by an extraction process using 96% ethanol solvent and then tested on the pathogenic microbial *Staphylococcus aureus*. The research results showed that all bamboo shoots were able to inhibit the activity of pathogenic microbes, resulting in the appearance of a clear zone. The diameter of the clear zone formed was measured vertically and horizontally (Figure 3).

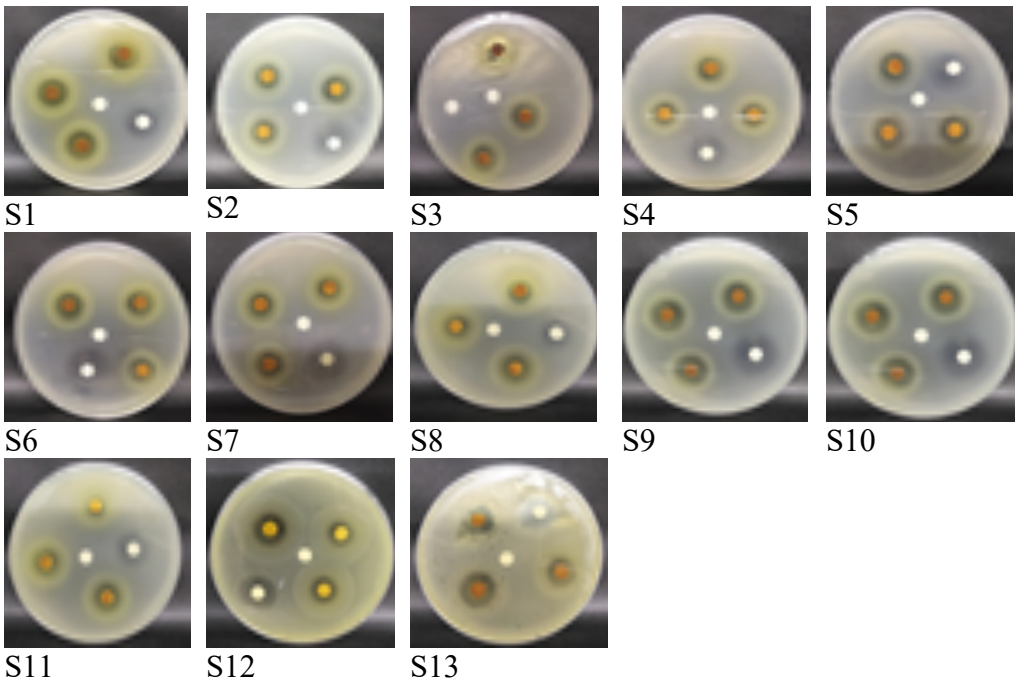


Figure 4. Morphology of bamboo shoot extract inhibited the pathogenic bacteria *Staphylococcus aureus*

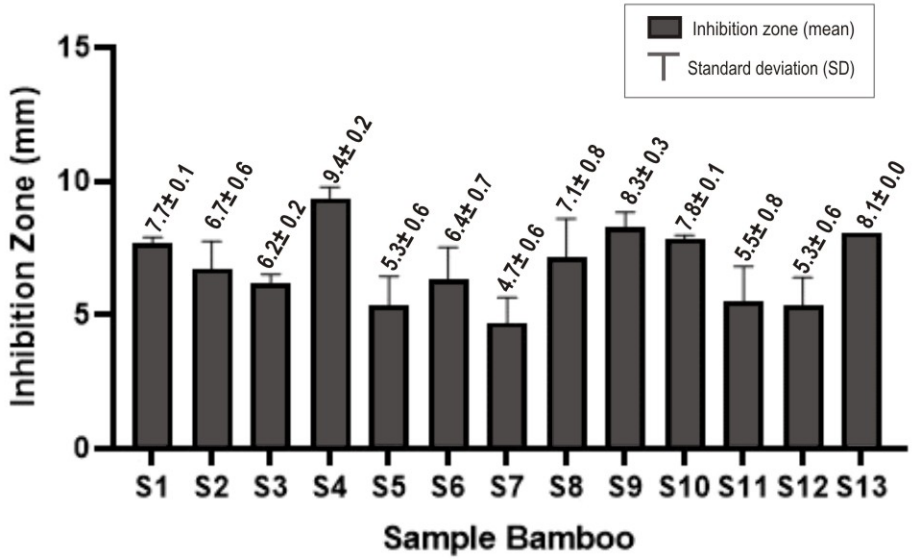


Figure 5. Antibacterial inhibition zone of 13 bamboo samples extracts from Bengkulu on the pathogenic microbial *Staphylococcus aureus*

Molecular character analysis

The genetic identification method for 13 bamboo species was based on the random applied polymorphic DNA (RAPD) method. The initial removal stage began with the selection of 81 primers, and 8 primers were successful in amplifying the bamboo DNA sequence, namely: OPA1, OPA8, OPA16, OPC11, OPD20, OPM4, OPM12, and OPM14. The visualization results of RAPD markers for 13 types of bamboo with 4 primers (Figure 6).

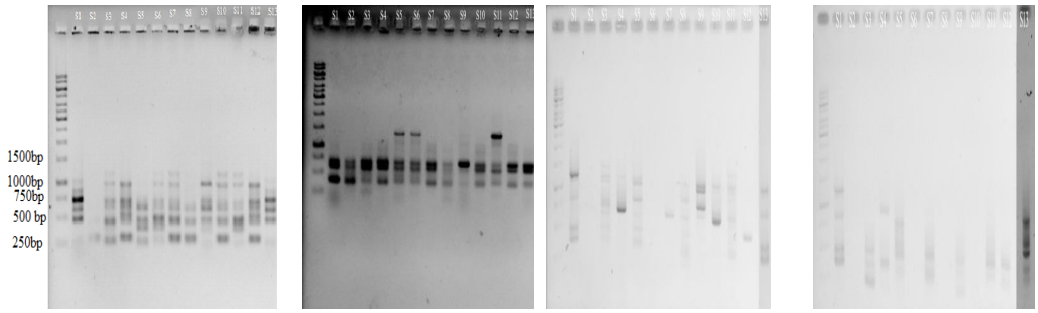


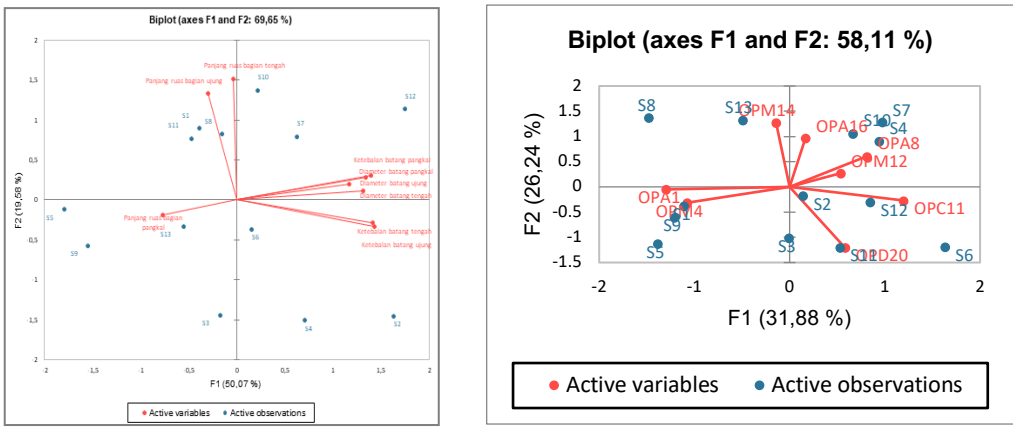
Figure 6. Visualization of RAPD markers on 13 Bengkulu bamboo species with primer (a) OPC11; (b) OPD20; (c) OPM4 and (d) OPM12

Table 3. The number and width of the amplification bands and the polymorphic results of 13 bamboos

Primer	Sequence (5' – 3')	Band size(bp)	Number of amplification bands	PIC
OPA 1	CAAT CGCC GT	291 – 958	21	1.41
OPA 8	GTGA CGTA GG	419 – 952	16	1.15
OPA 16	AGCC AGCG AA	339 – 920	13	1
OPC 11	AAAG CTGC GG	329 – 1326	66	3.72
OPD 20	ACCC GGTC AC	299 – 1371	29	2.33
OPM 4	GGCG GTTG TC	288 – 1323	32	1.97
OPM 12	GGGA CGTT GG	256 – 797	27	1.72
OPM 14	AGGG TCGT TC	348 – 1672	30	1.87
Total			**244**	

Principal component analysis (PCA)

Principal Component Analysis (PCA) is an analysis looking at the role of each morphological character in grouping (Franchi and Angulo, 2016). The results of PCA analysis of 13 species of bamboo showed that the morphological characteristics of bamboo showed a total diversity of 69.65% of the two main components; whereas based on molecular character, it was 58.11%. PCA analysis used 13 samples based on quantitative morphological characteristics consisting of 9 characters. Molecular properties used 8 different primers (Figure 7).



(a) (b)

Figure 7. Principal Component Analysis (PCA): (a) Morphological, (b) Molecular

Cluster analysis

the dendrogram graph of 13 bamboo samples based on morphological characters, it can be seen the similarity coefficient was 59%, divided into two clusters: the first cluster consists of S1, S8, S10, S12, S2 and S7; and the second cluster consists of S3, S4, S6, S11, S5, S9 and S13 (Figure 8). Based on molecular characters, it showed that the similarity coefficient was 79%, divided into three clusters: the first cluster consisting of S1, S2, S9, S6, S11, S5, S10, S3, S4 and S12; the second cluster consisting of S7 and S8; while the third cluster was S13.

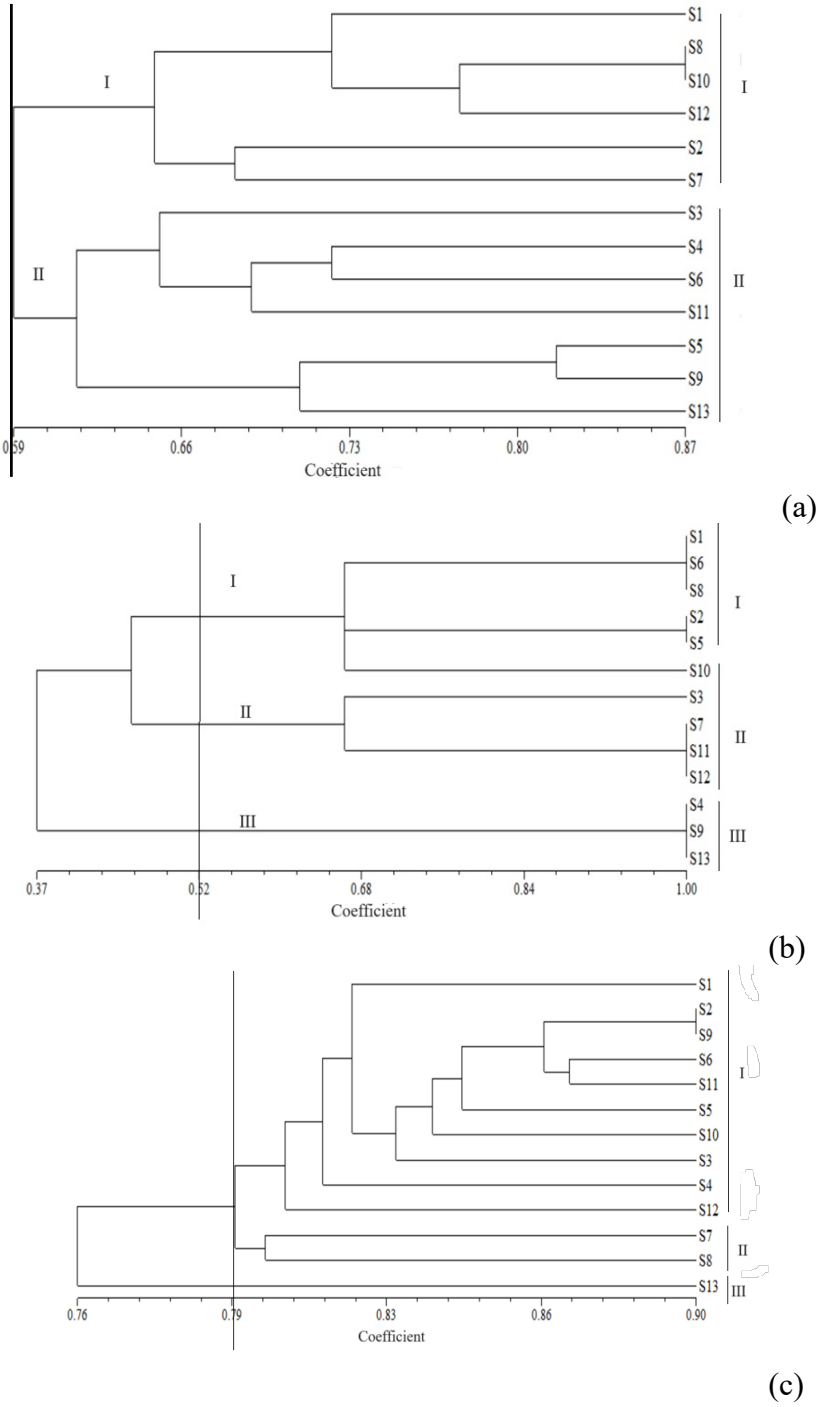


Figure 8. Dendrogram of 13 Bamboos Based on (a) Morphological (b) Antibacterial Characters c) Molecular Characters

Discussion

Morphological character analysis

Visual observation of bamboo stems showed that S1, S5, S8, S10, and S12 were close together and formed dense clumps; while bamboo S2, S3, S4, S5, S6, S7, S9, S11, and S13 did not form dense clumps. The surface of the nodes was smooth on bamboo S3, S4, S5, S9, and S13 and rough on S1, S2, S6, S7, S8, S10, S11, and S12. All bamboo stems had round segment shape and hollow segments, and the contents of the segments were dusty except for S7, where there was liquid found inside the chambers.

Some bamboos had hanging leaves, except for S2 bamboo with upright leaves and S7 bamboo with stiff leaves. All leaves were tapered in shape, with long leaves on bamboo S1, S2, S5, S7, S9, S10, S11, S12, and S13; while wide leaves were on S3, S4, S6, and S8. Rhizomes with short and thick diameter were found on bamboo S1, S2, S3, S4, S7, S8, S10; long and thin diameter were on bamboo S5, S6, S9, and S13; in addition, the position of the roots was particularly around the nodes.

The position of the branches found throughout the stem was on bamboos S1, S4, S7, S8, S10, and S12; branches at the top of the stem were on S2, S3, S5, S6, S9, S11, and S13. All bamboos had three branches with branch postures angled upwards, except bamboos S1and S3 were angled horizontally. Branches with thorns were found on S3, S7, S8, S10, and S13, but no thorns were seen on S1, S2, S4, S5, S6, S9, S11, and S13. The bamboo shoots collected were covered with feathers, except bamboo S3. The fur color was black and evenly distributed on S1, S2, S7, S9, and S10; the fur color was black and somewhat flat on S4, S5, S12, and S13; and the uneven brown fur were on S3, S6, S8, and S11.

Based on key morphological analysis, it can be seen that sample S1 is *Bambusa vulgaris Schrad var. striata*; S2, S7, S8, and S10 belong to *Bambusa vulgaris*; samples S3 and S13 are from species *Gigantochloa atter* kurz; S6 and S11 are *Gigantochloa serik*; samples S4 and S12 belong to *Dendrocalamus asper*; and samples S5 and S9 belong to *Schizostachyum* Kurz.

Based on the morphological findings, the identification key for the bamboo mentioned above is as follows:

1. a. smooth surface of the stem..... 2
   b. rough surface on the lower stem 3
2. a. all branch stems of the same dimension.....*Schizostachyum Kurz*
   b. one larger branch than the others .....*Dendrocalamus asper*
3. a. branches spread across the trunk 4
   b. branches at the top of the stem (middle segment) ..... *Gigantochloa serik*

4. a. shoots covered with black hairs 5
   b. shoots covered with brown hairs and wax ..... *Gigantochloa atter*
5. a. green reeds ..... *Bambusa vulgaris*
   b. green-striped, yellow reed ..... *Bambusa vulgaris var. striata*

The key morphological results above are in line with Widjaja *et al*. (2005), where Schizostachyum bamboo has branches of the same size, Dendroclamus asper shoots are coated with white wax. Bambusa vulgasis has green color and is upright, while Bambusa vulgaris striata has a yellow color with green lines.

A study by Chen (2020) found that the length of the 12 types of bamboo had different internode lengths, where the *Bambusa multiplex* bamboo type in Nanjing City is not much different from the length of the S3, S4, S6, S8, and S13 bamboo segments. The thickness of the stems of betung bamboo and ampel bamboo is not much different from S3, S4, S6, and S7 because the relationships tend to be closed, so they displayed the same phenotype (Park *et al*., 2021). Growth hormones and the environment influence the results of qualitative morphological observations. Growth hormones such as exogenous GA can affect the length of the internodes of Moso bamboo (*P. edulis*) and will affect plant height (Wu *et al*., 2023).

S5 and S9 have similar stem morphology where the distance between the stems does not form clumps, the nodes are dense, the surface of the nodes is not smooth, the surface of the internodes is rough below, the shape of the internodes is round, and the contents of the internodes are dusty. The distance between bamboo stalks forms a tight, dense, and close clump, but there are also types of bamboo that do not have dense clumps (Banik, 2015). Bamboo stems resemble round and cylindrical poles, have nodes, are hollow, have hard walls, and each node has a shoot or branch (Lorenzo and Mimendi, 2020). The cavity of the bamboo stem can be filled with water because the water absorption capacity of the bamboo plant is higher than that of other plants; most plants have an ability to absorb rainwater of 35%–40%, while bamboo is able to absorb 90% of the rainwater that falls, so the stem is rich in water content (Sofiah *et al*., 2018).

The nodes have a rough surface because they are covered by small aerial roots (Chen and Luo, 2020). Bamboo leaves have parallel bones like grass; each leaf has a prominent main leaf bone, which can cause the leaves to be stiff or upright. The tips of bamboo leaves are pointed, flat at the edges, lanceolate, and have a paper-like texture.

Rhizomes, or roots, appear on short bamboo segments. Bamboo roots with short rhizomes and thick necks will cause the roots to appear clustered; roots with short rhizomes and thinner necks will cause the clumps to grow more spread out (Banik, 2015). Bamboo stem branches are found on every stem segment, but

there are also bamboos that have stem branches only from the middle to the top of the stem. The position of the stem branches changes depending on the location of the buds. Several bamboo genera have branch growth starting from the middle to the top of the bamboo stem, namely *Dendrocalamus, Gigantochloa, and Schizostachyum* (Widjaja *et al*., 2005).

Variations in the number of bamboo branches were found to be greater than 1 branch. This is in line with research (Liu *et al*., 2020) on *Bambusa-Dendrocalamus-Gigantochloa complex* (BDG complex) that have more than 1 branch. In the *Gigantochloa, Dendrocalamus, and Bambusa genera*, one branch is larger than the other branches; whereas in the Schizostacyhun clan, the size of the branches is in the same size. There are branches that have thorns like thorny bamboo with a distinctive feature, namely the presence of thorns on the reeds and branches (Fathiya *et al*., 2022).

Bamboo shoots are generally covered in fine brown or black hairs (miang), and there is also bamboo shoots covered in wax. This is in line with Banik (2016) showing that betung bamboo is characterized by brown velvety hairs on the underside the reeds of the young ones, while the top is covered with white wax, which will disappear when old. In addition to such brown hair, bamboo feathers are also black, such as on S1, S2, S4, S5, S7, S9, S10, S12, and S13, similar to those of thorn bamboo and ampel bamboo.

Antibacterial character analysis

The diameter of the inhibition zone for 13 bamboo species can be seen in Figure 4. S4 (*Dendrocalamus asper* (Schult. and Schult.f) Backer) bamboo produces bamboo shoot extract with the highest inhibition zone, namely 9.4 ± 0.2 mm. Bamboo shoot extract from S7 (*Gigantochloa atter* (Hassk.) Kurz ex Munro) bamboo has the lowest inhibition zone, namely 4.7 ± 0.6 mm. In line with research on betung bamboo shoots (*Dendrocalamus asper*) at a concentration of 60% extracted in a 96% ethanol solution, it is known to be able to inhibit the pathogenic bacteria *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* with an inhibitory zone diameter of 9.05 ± 0.12 mm and 5.07 ± 0.13 mm, respectively (Artanti and Mujahidah, 2021).

Molecular character analysis

Based on the 8 primers used, genetic differences between bamboo genera and species could be generated. The results of this study are in line with Konzen *et al*. (2017) who proposed that byanalyzing bands with strong intensity, it is possible to differentiate genera and species genetically. Based on the research

results, it is known that the number of bands successfully amplified was 244, with the number of polymorphic bands being 244 and PIC having the lowest value of 1.00 and the highest value of 3.72. The more bands obtained at a locus, the better information about the polymerase is provided. The PIC value is shown to be higher than RAPD (Annisa *et al*., 2019). The PIC percent was known to be 1, r based on a comparison between the number of polymorphism bands and all the bands formed. The assumption that the PIC value of <0.3 which was not informative; 0.3–0.59 was informative; and > 0.6 was very informative. It means that in this study, the PIC value was very informative.

Principal component analysis (PCA)

S1, S8, and S11 showed similarities strongly influenced by the length of the middle and tip segments; S5 and S9 had similarities affected by the length of the base segments. So it can be seen that morphological characters are significant in plant diversity; this is in line with (Liana *et al*., 2017) that the length of the 5th segment can separate these plants from the *Schisoztachyum* genus. While the direction of the branches and the location of the branches can separate the plants of the genus Natus. Meanwhilethe ligula ear reeds separate the plants of the genus *Gigantochloa*, and the roots play the role of separating the genus *Dendrocalamus*.

The bamboo showed diversity, where bamboo with the closest similarity, including bamboo S3 and S11, was strongly influenced by the OPD 20 primer; S4, S7, and S10 were strongly influenced by primers OPA 8 and OPA 16; and S5, S9, and S1 were strongly influenced by the OPM 4 primer.

Cluster analysis

Samples S8 and S10 had the highest similarity coefficient based on morphological characters (87%), while samples S2 and S9 had the highest similarity coefficient based on molecular characters (90%). The greater the coefficient value, the higher the sample similarity (Lukmanasari *et al*. 2020). Morphology molecular are able to differentiate each into several groups; this is in line with Konzen *et al*. (2017), showing a coefficient of 42%. Thus, three groups were found, namely *Phyllostachs, Dendrocalamus,* and *Bambusa*. The similarity value is at least 82% for *Bambusa*; 92% for *Phyllostachys*; and 100% for *Dendrocalamus* for each genus considered within one group, which showing that RAPD is able to differentiate bamboo taxa.

In conclusion, based on the results above, it can be seen the diversity of bamboo types can be determined based on morphological, antibacterial and

molecular analyses. The key morphological analysis of the 13 samples studied consisted of six species and three genera of bamboo with two bamboo clusters with a similarity coefficient of 59%. Based on the molecular analysis, it consists of three clusters at a similarity coefficient of 79%. based on antibacterial analysis, it consists of 3 clusters with a similarity coefficient of 52%.

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References

Amom, T., Tikendra, L., Apana, N., Goutam, M., Sonia, P., Koijam, A. S., Potshangbam, A. M., Rahaman, H. and Nongdam, P. (2020). Efficiency of RAPD, ISSR, iPBS, SCoT and phytochemical markers in the genetic relationship study of five native and economical important bamboos of North-East India. Phytochemistry, 174(February), 112330. https://doi.org/10.1016/j.phytochem.2020.112330

Annisa, Hafzari, R., Setiawati, T., Irawan, B. and Kusmoro, J. (2019). Evaluation of RAPD markers for molecular identification of five bamboo genera from Indonesia. Folia Forestalia Polonica, Series A, 61:255-266. https://doi.org/10.2478/ffp-2019-0025

Artanti, A. N. and Mujahidah, F. S. (2021). Antibacterial activity test of ethanol extract and SAP of Betung Bamboo Shoot (*Dendrocalamus asper*) Against Klebsiella pneumoniae and Pseudomonas aeruginosa Bacteria. Journal of Biodiversity and Biotechnology, 1:11. https://doi.org/10.20961/jbb.v1i1.50519

Banik, R. L. (2015). Morphology and Growth. Springer International Publishing Switzerland. https://doi.org/10.1007/978-3-319-14133-6_3

Banik, R. L. (2016). Chapter 4 Dendrocalamus Nees In: Silviculture of South Asian Priority Bamboos. Springer Singapore. https://doi.org/10.1007/978-981-10-0569-5

Chen, G. and Luo, H. (2020). Effects of node with discontinuous hierarchical fibers on the tensile fracture behaviors of natural bamboo. Sustainable Materials and Technologies, 26, e00228. https://doi.org/10.1016/j.susmat.2020.e00228

Chen, X. (2020). Variations in patterns of internode and branch lengths for several bamboo species bamboo species. Plant Biosystems - An International Journal Dealing with All Aspects of Plant Biology, 0:1-12. https://doi.org/10.1080/11263504.2020.1829729

Damayanto, I. P. G. P. and Widjaja, E. A. (2017). A noteworthy Dendrocalamus (Poaceae: Bambusoideae) from Sumatra, Indonesia. Gardens' Bulletin Singapore, 69:75-80. https://doi.org/10.26492/gbs69(1).2017-04

de Jesus, O. N., de Freitas, J. P. X., Dantas, J. L. L. and de Oliveira, E. J. (2013). Use of morpho-agronomic traits and DNA profiling for classification of genetic diversity in papaya. Genetics and Molecular Research, 12:6646-6663. https://doi.org/10.4238/2013.July.11.8

Deshwal, R. P. S., Singh, R., Malik, K. and Randhawa, G. J. (2005). Assessment of genetic diversity and genetic relationships among 29 populations of Azadirachta indica A. Juss. using RAPD markers. Genetic Resources and Crop Evolution, 52:285-292. https://doi.org/10.1007/s10722-005-3282-5

Doyle, J. J. and Doyle, J. (1990). Isolation of plant DNA from fresh tissue. Focus, 12:13-15.

Ellegren, H. and Galtier, N. (2016). Determinants of genetic diversity. Nature Reviews Genetics, 17:422-433. https://doi.org/10.1038/nrg.2016.58

Ervianti, D., Widjaja, E. A. and Sedayu, A. (2019). Bamboo diversity of Sulawesi, Indonesia. Biodiversitas, 20:91-109. https://doi.org/10.13057/biodiv/d200112

Fathiya, N., Qariza, M. H., Nazhifah, S. A. and Diah, H. (2022). Morphological characteristics and utilization of thorny bamboo (*Bambusa blumea*) in the coastal area of Jambo Timu Village, Blang Mangat Subdistrict, Lhokseumawe. Jurnal Jeumpa, 9:767-776. https://doi.org/10.33059/jj.v9i2.6314

Feng, X., Tan, S., Dong, Y., Zhang, X., Xu, J., Zhong, L. and Yu, L. (2023). Mapping Large-Scale Bamboo Forest Based on Phenology and Morphology Features. Remote Sensing, 15:1-17. https://doi.org/10.3390/rs15020515

Franchi, G. and Angulo, J. (2016). Morphological principal component analysis for hyperspectral image analysis. ISPRS International Journal of Geo-Information, 5(6). https://doi.org/10.3390/ijgi5060083

Gu, H., Zhang, C., Wang, J., Shi, X., Xia, R., Liu, B., Chen, F. and Bu, W. (2019). Variation in basic morphological and functional traits of Chinese bamboo. Biodiversity Science, 27:585-594. https://doi.org/10.17520/biods.2019001

Hapsari, L., Agency, I., Wahyudi, D. T. and Azrianingsih, R. (2015). Genome identification of bananas (*Musa* L.) from East Java Indonesia assessed with PCR-RFLP of the Internal Transcribed Spacers Nuclear Ribosomal DNA. International Journal of Biosciences (IJB), 7:42-52. https://doi.org/10.12692/ijb/7.3.42-52

Hastuti, R. W., Primairyani, A. and Ansori, I. (2018). Studi Keanekaragaman Jenis Bambu Di Desa Tanjung Terdana Bengkulu Tengah. Diklabio: Jurnal Pendidikan Dan Pembelajaran Biologi, 2:96-102. https://doi.org/10.33369/diklabio.2.1.96-102

Konzen, E. R., Peron, R., Ito, M. A., Brondani, G. E. and Tsai, S. M. (2017). Molecular identification of bamboo genera and species based on RAPD-RFLP markers. Silva Fennica, 51(4). https://doi.org/10.14214/sf.1691

Kumalasari, R., Iwansyah, A. C., Ratnawat, L., Fitrianti, I. and Darmajana, D. A. (2019). Effect of pre-treatment on nutrient, antinutrient, and antioxidant properties of dried shoots from some edible Indonesian bamboo species. African Journal of Food, Agriculture, Nutrition and Development, 19:14932-14949.

Liana, A., Purnomo, P., Sumardi, I. and Daryono, B. S. (2017). Ethnobotany of Bamboo in Sangirese, North Celebes. Biosaintifika: Journal of Biology & Biology Education, 9:81. https://doi.org/10.15294/biosaintifika.v9i1.7405

Liu, J., Zhou, M., Yang, G., Zhang, Y., Ma, P. and Guo, C. (2020). Molecular phylogenetics and evolution ddRAD analyses reveal a credible phylogenetic relationship of the four main genera of Bambusa - Dendrocalamus - Gigantochloa complex (Poaceae : Bambusoideae). Molecular Phylogenetics and Evolution, 146:106758. https://doi.org/10.1016/j.ympev.2020.106758

Lorenzo, R. and Mimendi, L. (2020). Digitisation of bamboo culms for structural applications. Journal of Building Engineering, 29:101193. https://doi.org/10.1016/j.jobe.2020.101193

Lukmanasari, P., Purwantoro, A., Murti, R. H. and Zulkifli. (2020). Similarity Level of Nepenthes spp. Based on the Qualitative Character, 5:140-149. https://doi.org/https://doi.org/10.22146/ipas.55728

Maftuchah, Febriana, L., Sulistyawati, Reswari, H. A. and Septia, E. D. (2021). Morphological diversity and heritability of nine local sorghum (*Sorghum bicolor*) genotypes in east java, indonesia. Biodiversitas, 22:1310-1316. https://doi.org/10.13057/biodiv/d220330

Oggier, F. and Datta, A. (2023). On parsimony and clustering. PeerJ Computer Science, 1-17. https://doi.org/10.7717/peerj-cs.1339

Park, S., Lee, M., Febrianto, F. and Wistara, N. J. (2021). Effects on Morphology and Chemical Properties of Indonesian Bamboos by Carbonization. Jurnal Sylva Lestari, 9:190. https://doi.org/10.23960/jsl29190-201

Peronika, T., Suharyanto and Soetrisno, E. (2022). Introduksi Teknologi Lemea untuk Produk Daging Sapi dengan Lama Fermentasi yang Berbeda. Buletin Peternakan Tropis, 3:24-32. https://doi.org/10.31186/bpt.3.1.24-32

Probojati, R. T., Wahyudi, D. and Hapsari, L. (2019). Clustering analysis and genome inference of pisang raja local cultivars (*Musa* spp.) from Java Island by Random Amplified Polymorphic DNA (RAPD) Marker. Journal of Tropical Biodiversity and Biotechnology, 4:42-53. https://doi.org/10.22146/jtbb.44047

Ramanatha Rao, V. and Hodgkin, T. (2002). Genetic diversity and conservation and utilization of plant genetic resources. Plant Cell, Tissue and Organ Culture, 68:1-19. https://doi.org/10.1023/A:1013359015812

Reddy, G. T., Reddy, M. P. K., Lakshmanna, K., Kaluri, R., Rajput, D. S., Srivastava, G. and Baker, T. (2020). Analysis of Dimensionality Reduction Techniques on Big Data. IEEE Access, 8:54776-54788. https://doi.org/10.1109/ACCESS.2020.2980942

Ritonga, M. A., Syamsuardi, Nurainas and Damayanto, I. P. G. P. (2023). Bamboo diversity in Weh Island, Aceh, Indonesia. Biodiversitas, 24:2563-2576. https://doi.org/10.13057/biodiv/d240508

Sofiah, S., Setiadi, D. and Widyatmoko, D. (2018). The influence of edaphic factors on bamboo population in Mount Baung Nature Tourism Park , Pasuruan , East Java , Indonesia. Tropical Drylands, 2:12-17. https://doi.org/10.13057/tropdrylands/t020103

Wahidah, B. F., Damayanto, I. P. G. P. and Mulyani, S. (2021). Bamboo Diversity in Indrokilo Botanical Garden, Central Java. Buletin Plasma Nutfah, 27:57. https://doi.org/10.21082/blpn.v27n1.2021.p57-70

Widjaja, E. A., Astuti, I. P., Arinasa, I. B. K. and Sumantera, I. W. (2005). Identikit Bambu di Bali (Issue January 2005). Bidang Botani Puslit Biologi-LIPI.

Widjaja, E. A., Ervianti, D. and Kusumaningtyas, H. (2020). *Buku Saku Identitas Bambu*. Direktorat Inventarisasi dan Pemantauan Sumber Daya Hutan.

Wu, C., Bai, Y., Cao, Z., Xu, J., Xie, Y., Zheng, H., Jiang, J., Mu, C., Cheng, W., Fang, H. and Gao, J. (2023). Plasticity in the morphology of growing bamboo: A bayesian analysis of exogenous treatment effects on plant height, internode length, and internode numbers. Plants, 12(8). https://doi.org/10.3390/plants12081713

Yadav, M. and Mathur, A. (2021). Bamboo as a sustainable material in the construction industry: An overview. Materials Today: Proceedings, 43:2872-2876. https://doi.org/10.1016/j.matpr.2021.01.125

Yani, A. P. (2012). Keanekaragaman Dan Populasi Bambu Di Desa Talang Pauh Bengkulu Tengah. Exacta, 10:61-70.

Yani, A. P. and Anggraini, N. (2018). Peranan Bambu Dalam Kehidupan Masyarakat Desa Taba Terunjam Bengkulu. Prosiding Seminar Nasional Pendidikan Biologi, 924-928.

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