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Research Article

Morphological Characterization and Phenetic Relationships of *Schizophyllum commune* Fr. from Diverse Host Plants

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ABSTRACT

Schizophyllum commune is a cosmopolitan basidiomycetous fungus that plays an important role in wood decomposition and nutrient cycling, and is increasingly recognized as a plant pathogen. This study characterized *S. commune* from diverse host plants across Java Island, Indonesia, using integrated morphological, molecular, and phenetic approaches. Fruiting bodies were collected from 16 locations in West Java, Central Java, East Java, and the Special Region of Yogyakarta, representing nine host plants with *Swietenia mahagoni*, *Albizia chinensis* and *Rubus idaeus,* to our knowledge, were never documented as host plants of *S. commune.* Morphological characterization based on 35 macroscopic and microscopic characters revealed three pileus shape types which were orbicular, suborbicular, and flabelliform with the flabelliform type being the most frequent across hosts. Molecular identification using ITS rDNA sequencing confirmed that all representative isolates from the three pileus groups belonged to *S. commune,* showing high sequence similarity (≥99%) with Genbank reference strains. Phenetic analysis using UPGMA grouped the isolates into two major clusters with high similarity values (0.66-1.00), indicating close relationships among isolates from different regions and host plants. Principal component analysis showed that pileus morphology and fruit body attachment characteristics were the main contributors to intraspecific variation.

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1. INTRODUCTION

Schizophyllum commune is a globally widespread basidiomycetous fungus within the phylum Basidiomycota and the class Agaricomycetes. It is commonly referred to as the split-gill fungus due to the characteristic longitudinally divided gill-like folds on the underside of its fruiting body, a morphological trait that facilitates species identification The basidiocarps are typically small, fan-shaped, and sessile on substrates such as dead wood (Padhiar et al., 2009). As a predominantly saprotrophic organism, *S. commune* colonizes decaying lignocellulosic material and produces an extensive suite of carbohydrate-active enzymes that contribute to the breakdown of cellulose, hemicellulose, and pectin, thereby playing a significant role in nutrient cycling and wood decomposition in forest ecosystems (Zhu et al., 2016).

S. commune is capable of colonizing dead woody material from more than 150 plant genera, although it is most frequently encountered on fallen branches and logs of hardwood species. The fungus is able to decompose all major plant cell-wall components, a capacity supported by a genome enriched in genes encoding pectinases, hemicellulases, and cellulases (Ohm et al., 2010). Cooke (1961) originally characterized *S. commune* as a wound parasite or, alternatively, a saprotroph. Consistent with this view, the fungus is frequently observed colonizing dead branches on otherwise living trees (Takemoto et al., 2010). However, *S. commune* is now widely recognized as a plant pathogen. It is known to cause wood rot in a range of living tree hosts, including both fruit and ornamental species (Oprea et al., 1994; Dikin et al., 2003). Based on field observations in Central Sulawesi, Indonesia, 92 types of weathered wood representing 36 plant families were identified as substrates supporting the growth of *S. commune* (Yusran et al., 2023). Whereas Dasanayaka & Wijeyaratne (2017) evaluated seven wood types as sawdust substrates for the cultivation of *S. commune*, and found that the highest yield was observed on jackfruit (*Artocarpus heterophyllus*) sawdust and the lowest on thungfaa (*Alstonia macrophylla*) sawdust.

The morphological characteristics of *S. commune* are central to understanding its biology, taxonomy, ecology, and practical applications. Observations at both macroscopic and microscopic levels provide critical information for accurate species identification within the Basidiomycota, many of which share similar wood-decaying niches (Robledo et al., 2014; Dasgupta, 2025). Distinctive features such as fan-shaped basidiomata and longitudinally split gills serve as reliable diagnostic markers for field and laboratory identification, minimizing ambiguity in species recognition and facilitating ecological surveys (Takemoto et al., 2010). Kusrinah and Kasiamdari (2015) documented the macroscopic and microscopic morphology of *S. commune* collected from various regions in Java, showed the fruit body morphology varied between young and mature phases, while microscopic features remained largely consistent across samples. The morphological characters such as cap shape, surface texture, and hymenial structures could be used to describe growth phases of the fungus, contributing to detailed species characterization. The environmental factors and geographic separation contribute to morphological diversity in *S. commune*, while overall phenetic similarity supports its classification as a single species. The study highlights the usefulness of morphological data in assessing phenetic relationships, although molecular approaches are needed to improve the accuracy of taxonomic identification.

Molecular identification complements morphological analysis, providing more reliable identification of *S. commune*. Molecular identification of *S. commune* is commonly conducted using sequencing of the internal transcribed spacer (ITS) region of ribosomal DNA, which is widely accepted as the universal DNA barcode for fungi (White et al., 1990). The ITS region, consisting of ITS1, 5.8S rRNA, and ITS2, exhibits high interspecific variability while remaining conserved within species, enabling accurate discrimination of *S. commune* from closely related fungal taxa (Schoch et al., 2012). Several studies have demonstrated that ITS sequencing effectively confirms morphological identification of *S. commune*, particularly when morphological characteristics are insufficient or ambiguous, such as in environmental isolates (Chowdhary et al., 2013). Comparative analyses of ITS sequences using BLAST searches against GenBank databases consistently show high sequence similarity (≥98-100%) among *S. commune* isolates, supporting the reliability of ITS-based identification and phylogenetic placement (Buzina et al., 2001).

The morphology of *S. commune* fruiting bodies can vary significantly depending on the wood substrate. Field observations have shown that the fungus produces larger, grayish basidiocarps on *Lannea coromandelica* compared with smaller, whiter fruiting bodies on *Corypha* wood, demonstrating substrate-related differences in macroscopic traits. Similarly, cultivation experiments on sawdust from seven different tree species revealed that substrate type affects growth, yield, and the physical characteristics of the fruiting bodies (Dasanayaka & Wijeyaratne, 2017). These results suggest that both the type of wood and its nutrient composition play key roles in shaping the observable morphology of *S. commune* under natural and experimental conditions. This study aims to characterize and identify *S.*

commune from diverse host plants using morphological and molecular approaches and to determine its phenetic relationship based on morphological data.

2. METHODOLOGY

2.1. Sample Collection

S. commune fruiting bodies were collected from 16 locations of West Java East Java, Central Java, and the Special Region of Yogyakarta (Figure 1). Geographically, West Java Province is located between approximately 5°50′ and 7°50′ South Latitude and 104°48′ and 108°48′ East Longitude, Central Java Province is located between 6° and 8° South Latitude and between 108° and 111° East Longitude, East Java Province is located between approximately 7°12′ and 8°48′ South Latitude and 111°00′ and 114°04′ East Longitude, while the Special Region of Yogyakarta is situated in the southern part of Java Island and lies approximately between 7°48′ and 8°12′ South Latitude and 110°05′ and 110°42′ East Longitude. Java Island has a tropical monsoon climate with a wet season from around October to April, during which rainfall is generally moderate to high, often reaching about 250-500 mm per month in many areas (BMKG, 2024).

Figure 1. *S. commune* sampling collection in West Java (A), Central Java (B), West Java (C) and Special Region of Yogyakarta (D), Indonesia

S. commune samples were collected from decaying wood in their natural environment. Fresh basidiocarps were carefully removed using sterile instruments and placed in clean, labelled containers to minimize contamination and damage during transport. The specimens were subsequently transported to the laboratory for detailed morphological observation and further analyses. Only healthy, fully developed fruiting bodies were selected to ensure accurate morphological characterization and high-quality DNA for molecular identification.

2.2. Morphological Characterization

The observation of *S. commune* samples were made on 35 qualitative and quantitative characters following the references from Carreno-Ruiz et al. (2019), Cooke (1961), Dawson et al. (2018), and Putra (2021) (Table 1). Morphological characterization of *S. commune* was performed using macroscopic and microscopic observations in accordance with standard mycological procedures. Macroscopic features of fresh fruiting bodies, including pileus shape and size, colour, surface texture, lamellae structure, stipe presence, growth habit, and fruiting body consistency, were recorded. Microscopic observations were carried out on prepared slides from fresh specimens and cultured material to examine spores, hyphae, cystidia, and clamp connections using a light microscope.

Table 1. Quantitative and Qualitative Characteristics in *S. commune*

Character number	Parameter
1	Life strategies (saprophyte, endophyte)
2	Growth type (solitary, scattered)
3	Fruit body attachment type (eccentric, central)
4	Fruit body type (agaroid, resupinate corticioid, discomycetoid, pileate corticioid, pileate polyporoid, resupinate polyporoid, ramarioid, stromatoid, tremelloid)
5	Fruit body colour (white, beige, dark beige)
6	Fruit body structure (annual, perennial)
7	Pileus shape (flabelliform, suborbicular, orbicular)
8	Pileus surface structure (glabrous, hairy, scaly, tomentum, trichoderm)
9	Pileus size (measurement length x width in cm)
10	Upper top pileus (dry, slimy)
11	Pileus status (simple, imbricate)
12	Pileus margin (plane, decurved, crenate, irregular, lobbed, ragged)
13	Pileus texture (tough, soft)
14	Pileus diameter (measurement in cm)
15	Hairy upper pileus (absent, present)
16	Mitic system (monomitic, dimitic, trimitic)
17	Hymenophore type (smooth, poroid, labyrinthine , lamellate, denticulate, gasteroid, irregular)
18	Hymenophore surface (presence of sterile structures, absent of sterile structure)
19	Density lamella (narrow, intermediate, distant)
20	Spore type (conidia, oidia, chlamydospores, basidiospore)
21	Spore shape (cylindric, allantoid, lunate, navicular, oblong ellipsoid)
22	Spore ornamentation (reticulose, russuloid, piny, verrucose, rugose)
23	Spore colour (hyaline, yellow brown)
24	Spore size (measurement length x width in µm)
25	Pellicle (absent, present)
26	Pellicle colour (white, light gray, dark gray, light yellow, light brown)
27	Context thickness (measurement in cm)
28	Hymenium length (measurement in cm)
29	Hymenium colour (white, light gray, dark gray, light yellow, light brown)
30	Abhymenial hair constitution (measurement in cm)
31	Abhymenial hair colour (white, light gray, dark gray, light yellow, light brown)
32	Clamp connection (absent, present)
33	Hyphae (septate, aseptate)
34	Cystidia (absent, present)
35	Chlamydospore (absent, present)

2.3. Molecular Identification

Genomic DNA was extracted with DNA extraction Kit, Tiangen Biotech, Beijing China according to the procedure in the kit with some modifications as follows: Fresh mycelia taken from 10-day-old cultures were frozen with liquid nitrogen. Frozen mycelia were ground with a sterilized mortar and stored in 1.5 mL microtubes. A total of 500 µL of extraction buffer was added to each microtube and incubated at 65°C for 30 min. After incubation, the samples were vortexed and then centrifuged at 4°C for 10 min at 12,000 rpm. After that, they were transferred to 1.5 mL microtubes and added with 1000 µL of 99.9% alcohol and then centrifuged at 4°C for 5 min at 12,000 rpm. Then, the supernatant was discarded and 500 µL of 70% alcohol was added. The precipitated DNA was then vortexed and centrifuged at 4°C for 5 min at 12,000 rpm. The supernatant was removed and the remaining alcohol was evaporated. The DNA was resuspended in 500 µL of sterile distilled water. DNA concentration was measured using a spectrophotometer (Alam et al., 2010).

The polymerase chain reaction (PCR) was performed using universal primers ITS1 (5′-TCC GTA GGT GAA CCT GCG G-3′) and ITS4 (5′-TCC TCC GCT TAT TGA TAT GC-3′) (White et al., 1990). The amplification reaction was carried out in a total volume of 20 µL containing PCR buffer (10x, $MgCl_2$ 25 mM, dNTPs 10 mM, ITS primers 100 pmols and *Taq* DNA Polymerase 5 U/µL, DNA 10 ng/µL. PCR reaction was performed on a GeneAmp PCR System 9700 (Applied Biosystems, Foster City, CA) with

an initial denaturation step of 5 min at 95°C, followed by 35 cycles of denaturation for 30 sec at 94°C, annealing for 30 sec at 52°C, extension for 1 min at 72°C, and a final extension for 10 min at 72°C. The amplification products were analyzed by gel electrophoresis on a 1.5% agarose gel with 1 kb DNA ladder as a marker.

2.4. Phenetic Relationships

The phenetic relationships of *S. commune* were analysed based on 35 morphological characters. Each observed character was scored and then converted into binary data for analysis. A similarity matrix was calculated using the Gower general similarity coefficient, and phenetic relationships were examined through cluster analysis using the Unweighted Pair Group Method with Arithmetic Averages (UPGMA). The results of cluster analysis were constructed in dendrogram. Principal Component Analysis (PCA) was also performed based on variation found on the morphological characters that contributed to distinguish each cluster. Both the dendrogram and PCA analyses were conducted using Multivariate Statistical Package (MVSP) software, version 3.2

3. RESULTS AND DISCUSSION

3.1. Sample Collection

Sampling was conducted across Java Island, including West Java, East Java, Central Java, and Special Region of Yogyakarta. Table 2 summarizes the sampling locations and the different host trees from which *S. commune* was collected in this study.

Table 2. Collected Samples of *S. commune* from Diverse Host Plants

Province	District	Host Plant		Sample code
		Scientific name	Common name	
West Java	Cirebon	*Mangifera indica* L.	Mango	SCMg
	Cirebon	*Leucaena leucocephala* (Lam.) de Wit.	Leucaena	SCPc
	Kuningan	*Rubus idaeus* L.	Red Raspberry	SKAr
	Kuningan	*Mangifera indica* L.	Mango	SKMg
	Kuningan	*Swietenia mahagoni* (L.) Jacq.	Mahogany	SKMh
	Kuningan	*Tamarindus indica* L.	Tamarind	SKAs
East Java	Sidoarjo	*Saccharum officinarum* L.	Sugarcane	SSTb
	Sidoarjo	*Mangifera indica* L.	Mango	SSMg
	Sidoarjo	*Lannea coromandelica* (Houtt.) Merr.	Ash Tree	SSJr
	Malang	*Mangifera indica* L.	Mango	SMMg
Central Java	Semarang	*Tectona grandis* L. f.	Teak	SSJt
	Semarang	*Leucaena leucocephala* (Lam.) de Wit	Leucaena	SSPc
	Semarang	*Albizia chinensis* (Osbeck) Merr	Albizia	SSsg
	Ambarawa	*Albizia chinensis* (Osbeck) Merr.	Albizia	SASg
	Temanggung	*Swietenia mahagoni* (L.) Jacq.	Mahogany	STMh
Special Region of Yogyakarta	Sleman	*Albizia chinensis* (Osbeck) Merr.	Albizia	SYSg

Table 2 summarizes the collection of *S. commune* samples across several provinces in Java, Indonesia, highlighting the diversity of host plants and sampling locations. In West Java, samples were obtained from Cirebon and Kuningan districts, with hosts including *Mangifera indica* (Mango), *Leucaena leucocephala* (Leucaena), *Rubus idaeus* (Red Raspberry), *Swietenia mahagoni* (Mahogany), and *Tamarindus indica* (Tamarind). In East Java, samples were collected from Sidoarjo and Malang, with hosts including *Saccharum officinarum* (Sugarcane), Mango, and *Lannea coromandelica* (Ash Tree). In Central Java and Special Region of Yogyakarta, sampling focused on Semarang, Ambarawa, Temanggung, and Sleman, where the fungus was isolated from *Tectona grandis* (Teak), *Leucaena leucocephala*, *Albizia chinensis*, and *Swietenia mahagoni*.

The sample collection shows that *S. commune* is widely distributed across Java and can colonize nine host plants, including timber species, fruit trees, and crops. Its repeated occurrence on Mango (*Mangifera indica*) in West Java and Albizia (*Albizia chinensis*) in Central Java and Yogyakarta suggests frequent associations with these hosts. In East Java, it was also found on cultivated crops like sugarcane, highlighting its ecological versatility. These findings align with previous reports describing *S. commune* as a cosmopolitan, generalist basidiomycete capable of growing saprophytically on decaying wood or as an opportunistic parasite on stressed or wounded living trees (Kleijburg & Wösten, 2025).

Published research supports the observation of a diverse host range. In Mexico, *S. commune* has been reported on over 300 host species, indicating its wide adaptability to different substrates and climates beyond the usual woody habitats (Vázquez-Mendoza, 2013). Similarly, in field studies along the Palu-Koro fault in Central Sulawesi, *S. commune* was found growing on at least 92 different types

of weathered wood, further supporting its capacity to exploit numerous hosts (Yusran et al., 2023). These findings align with our results in Java, where the fungus was found on both hardwood trees (teak, mahogany) and agricultural crops (mango, sugarcane), suggesting no strict host specificity but rather a broad lignicolous and opportunistic ecology. However, to our knowledge, three previously unreported wood substrates were identified among the collected samples: *Swietenia mahagoni*, *Albizia chinensis*, and *Rubus idaeus.* The identification of these substrates expands the documented range of plant hosts and supports the view that *S. commune* exhibits broad host adaptability and ability to exploit diverse lignocellulosic resources in nature (Takemoto et al., 2010).

3.2. Morphological Characterization

The morphological analysis of *S. commune* revealed three distinct pileus shapes: orbicular, suborbicular, and flabelliform (Table 3, Figure 2). The orbicular shape was observed in samples SKMg, SSMg, and SYSg, collected from Mango (*Mangifera indica*) and Albizia (*Albizia chinensis*) substrates. The suborbicular shape was recorded in SSJt and STMh, which were obtained from Teak (*Tectona grandis*) and Mahogany (*Swietenia mahagoni*). The flabelliform shape was the most common, appearing in multiple samples (SCMg, SCPc, SKAr, SKMh, SKAs, SSPc, SSsg, SASg, SSTb, SSJr, SSMg) across a wide range of hosts, including Mango, Leucaena (*Leucaena leucocephala*), Red Raspberry (*Rubus idaeus*), Mahogany, Tamarind (*Tamarindus indica*), Albizia, Sugarcane (*Saccharum officinarum*), and Ash Tree (*Lannea coromandelica*).

Table 3. Groups of morphological characteristics based on the pileus shape of *S. commune*

Pileus shape	Sample code	Host Plant
Orbicular	SKMg, SSMg, SYSg	Mango, Albizia
Suborbicular	SSJt, STMh	Teak, Mahogany
Flabelliform	SCMg, SCPc, SKAr, SKMh, SKAs, SSPc, SSsg, SASg, SSTb, SSJr, SMMg	Mango, Leucaena, Red Raspberry, Mahogany, Tamarind, Albizia, Sugarcane, Ash Tree

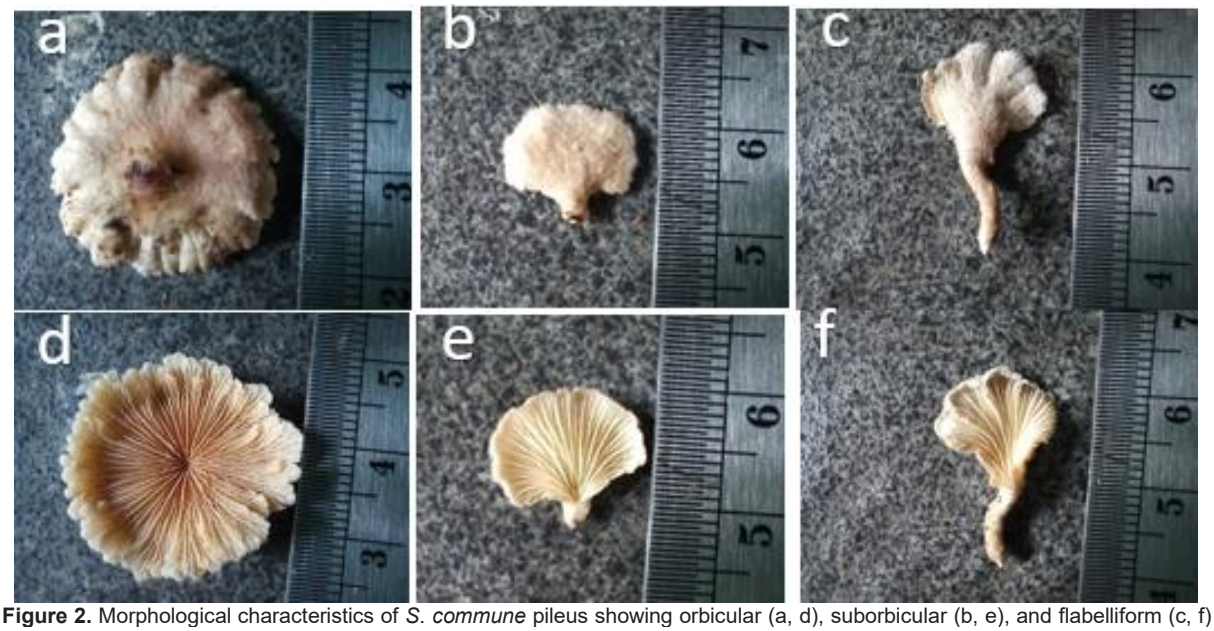

Figure 2. Morphological characteristics of *S. commune* pileus showing orbicular (a, d), suborbicular (b, e), and flabelliform (c, f) shapes. a, b, c = upper surface; d, e, f = lower surface

The observed variation in pileus morphology aligns with previous studies showing that *Schyzophyllum commune* exhibits considerable phenotypic plasticity, influenced by factors such as environmental conditions, host substrate, and developmental stage. The widespread occurrence of flabelliform forms across multiple hosts suggests that this shape may be an adaptive feature for colonizing diverse lignocellulosic substrates, reflecting the species' generalist and opportunistic lifestyle (Yusran et al., 2023). This morphological diversity highlights the need to integrate macroscopic traits with molecular markers, such as ITS sequences, for accurate identification and characterization of *S. commune* populations in varying ecological settings (Liang et al., 2024).

Table 4 showed that *S. commune* exhibited three distinct pileus shapes groups which were orbicular, suborbicular, and flabelliform while most macroscopic and microscopic features remained consistent across groups. All were saprophytic, produced annual agaricoid fruit bodies, and had hairy, dry pilei with decurved margins. Growth varied slightly, with orbicular and flabelliform types scattered

and suborbicular types solitary. Fruit body color differed among groups, ranging from dark beige in orbicular, white to beige in suborbicular, and white, beige, or dark beige in flabelliform types. Microscopically, all groups shared a monomitic mycelial system, lamellate hymenophores with sterile structures, elliptical hyaline basidiospores (3.5-4.3 × 1-1.4 µm in length x width), white pellicles, septate hyphae, and clamp connections; cystidia and chlamydospores were not observed/absent.

Table 4. Morphological characters of *S. commune* based on pileus groups

Parameter	Group 1 (*orbicular*)	Group 2 (*suborbicular*)	Group 3 (*flabelliform*)
Life strategies	saprophyte	saprophyte	saprophyte
Growth type	solitary, scattered	solitary	scattered
Fruit body attachment type	central	eccentric	eccentric
Fruit body type	agaricoid	agaricoid	agaricoid
Fruit body colour	dark beige	white	white, beige, dark beige
Fruit body structure	annual	annual	annual
Pileus shape	orbicular	suborbicular	flabelliform
Pileus surface structure	hairy	hairy	hairy
Pileus size	0.65-2.5 x 1.3-2.5 cm	2.2-2.5 x 1.0-2.7 cm	0.7-2.5 x 1-3 cm
Upper top pileus	dry	dry	dry
Pileus status	simple	simple	simple, imbricate
Pileus margin	decurved, irreguler	decurved	decurved, irreguler, crenate
Pileus texture	soft	soft	tough, soft
Pileus diameter	1.3-2.5 cm	1.0-2.7 cm	1-3.2 cm
Hairy upper pileus	present	present	present
Mitic system	monomitic	monomitic	monomitic
Hymenophore type	lamellate	lamellate	lamellate
Hymenophore surface	presence of sterile structures	presence of sterile structures	presence of sterile structures
Density lamella	narrow	narrow	narrow
Spore type	basidiospore	basidiospore	basidiospore
Spore shape	elliptical	elliptical	elliptical
Spore ornamentation	absent	absent	absent
Spore colour	hyaline	hyaline	hyaline
Spore size (n=30)	3.5-4.3 x 1-1.4 µm	3.5-4.3 x 1-1.4 µm	3.5-4.3 x 1-1.4 µm
Pellicle	present	present	present
Pellicle colour	white	white	white
Context thickness	306-368 µm	100-363 µm	150-590 µm
Hymenium length	30 – 60.28 µm	14.04-24.08 µm	14.3 – 71.36 µm
Hymenium colour	white	white	white
Abhymenial hair constitution	thin	thin	thin
Abhymenial hair colour	white	white	white
Clamp connection	present	present	present
Hyphae	septate	septate	septate
Cystidia	absent	absent	absent
Chlamydospore	absent	absent	absent

Note: Bold characters are characters that show variation between samples and used for PCA analysis

The morphological analysis of *S. commune* showed three pileus shapes orbicular, suborbicular, and flabelliform while other macroscopic and microscopic traits remained consistent. All isolates were saprophytic, producing annual agaricoid fruit bodies with hairy, dry pilei and decurved margins, in line with previous reports (Liang et al., 2024). Growth patterns differed slightly, with orbicular and flabelliform types scattered and suborbicular types solitary. Fruit body colour varied by pileus type, ranging from dark beige in orbicular, white to beige in suborbicular, and multiple shades in flabelliform forms, reflecting substrate and environmental influences, as similarly observed in other studies of *S. commune* (Kleijburg & Wösten, 2025).

3.3. Molecular Identification

Molecular identification of the *S. commune* isolates was based on their pileus shape groups, with representative isolates being SKMg (orbicular group), STMh (suborbicular group), and SKAr (flabelliform group). Species identification was confirmed through DNA barcoding. PCR amplification of total fungal DNA using ITS1 and ITS4 primers produced single bands of 623 bp (SKMg, GenBank Acc.No. PZ240299), 642 bp (STMh, GenBank Acc.No. PZ241948), and 641 bp (SKAr, GenBank Acc.No. PZ240432). The amplified products were directly sequenced using the same primers, and the resulting ITS rDNA sequences were compared with corresponding regions of *S. commune* isolates available in GenBank. Homology searches revealed high similarity with strains from other countries. SKMg matched the Indian isolate SC/UK/02/2020 (MT909559) with 99% query coverage and 99.51% identity, STMh aligned with the French voucher 09-540 (MZ997086) with 100% coverage and 99.38% identity, and SKAr also matched the Indian isolate (MT909559) with 99% coverage and 99.84% identity (Table 5).

Table 5. Homology search of *S. commune* isolates with available sequences in GenBank

Isolates Code/ Accession Number	Highest Similarity	Accession	Country	Query Coverage	Percent Identity
SKMg/ PZ240299	*S. commune* isolate SC/UK/02/2020	MT909559	India	99%	99.51%
STMh/ PZ241948	*S. commune* voucher barcode 09-540	MZ997086	France	100%	99,38%
SKAr/ PZ240432	*S. commune* isolate SC/UK/02/2020	MT909559	India	99%	99.84%

The ITS sequence analysis confirmed that the isolates in this study are *S. commune*, showing strong similarity (>99%) to reference sequences from India and France, reflecting the species' global distribution. High sequence identity among the isolates and GenBank references highlights the low divergence of conserved ribosomal regions, consistent with previous reports that ITS and related rDNA regions reliably identify *S. commune* at the species level (>98-99% homology) (Choi et al., 2020). Similar studies of wild *S. commune* strains from diverse regions also reported ITS homologies of 99-100%, indicating limited variation despite geographic separation, a pattern corroborated by the close clustering of our isolates with GenBank sequences (Alam et al., 2010).

3.4. Phenetic Relationships

The dendrogram of *S. commune* isolates from Java, constructed using UPGMA based on morphological data, revealed the presence of two major clusters (Figure 3). The first cluster (cluster I) included SYSg, SSTb, SSJr, STMh, and SSPc, and SSJt while the second cluster (cluster II) comprised the remaining isolates, with SASg, SSsg, SKMh, SKMg, SKAr, SCPC.SMMg, SSMg, SKAs and SCMg forming a tightly linked subcluster. Similarity coefficients ranged from 0.82 to 1.0, indicating that several isolates from different locations share high morphological similarity, consistent with the ITS sequence homology results.

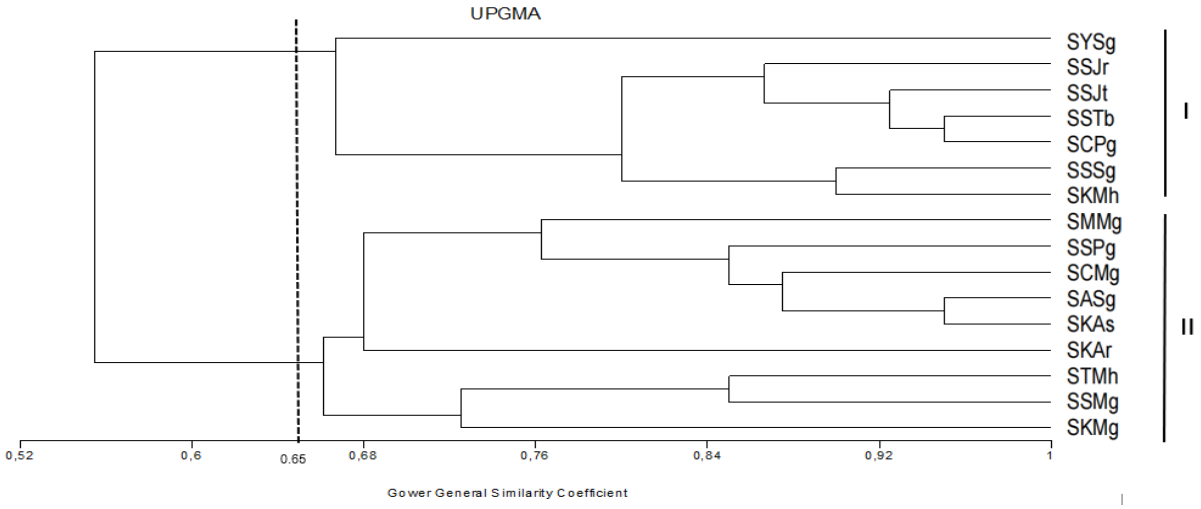

Figure 3. Dendrogram of *S. commune* from Java based on morphological data

Based on the phenetic relationship, the similarity values (0.66-1.0) indicate that while most isolates are very similar, some differences still exist, which is common in fungal populations. A phenon line at 65% indicates that the clusters formed share at least 65% similarity in characters, indicating moderate similarity in their characters based on the similarity index used (Sneath, 1973). Studies of *Pleurotus* mushrooms using UPGMA also found that isolates could be highly similar but still form distinct clusters (Khan et al., 2011). The morphological characters of *S. commune* isolates appeared generally similar among the sampled regions suggesting relatively limited morphological variation. The dendrogram demonstrates that while overall variation exists among the isolates, distinct subgroups can be identified based on shared morphological characters. The similarity coefficient at which the main split occurred in 65%. The clustering did not correspond to pileus shape categories and did not correspond to geographic origin. Based on the PCA analysis, there were 11 diagnostic characters that drive cluster separation. The high similarity values within subclusters indicate that several isolates, even from different geographic locations, share comparable morphological characteristics. This pattern is consistent with previous findings by Kusrinah & Kasiamdari (2015), who reported that *S. commune*

isolates from Java could be divided into two major groups based on morphological relationship analysis. PCA was performed to identify the main morphological characters contributing to variation among *S. commune* samples based on the data of Table 4 which showed 11 characters in bold (Table 6).

Table 6. PCA of morphological characters in *S. commune* isolates

Morphological character	PC1	PC2
Growth type	**-0,325**	-0,054
Fruit body attachment type	-0,196	**0,447**
Fruit body colour	-0,139	**0,442**
Pileus shape	**0,222**	**-0,454**
Pileus size	**0,504**	**0,246**
Pileus margin	-0,012	0,185
Pileus texture	**0,217**	0,047
Pileus status	-0,195	**0,388**
Pileus diameter	**0,504**	**0,246**
Context thickness	**-0,391**	0,02
Hymenium length	**-0,203**	**-0,296**
Eigen values	2,845	2,386
Percentage	25,862	21,693

Note: Bold character indicates main contributing character that distinguish between *S. commune*

The PCA of morphological characters in *S. commune* shows that the first two principal components explain 47.56% of the total variation, with PC1 accounting for 25.86% (Eigen value = 2.845) and PC2 accounting for 21.69% (Eigen value = 2.386). PC1 is mainly influenced by characters related to fruit body size, particularly pileus size and pileus diameter, indicating that variation along this axis reflects differences in basidiocarp dimensions among specimens. In contrast, PC2 is primarily associated with pileus shape, fruit body attachment type, fruit body colour, pileus status, suggesting that this component represents variation in the external morphology and attachment characteristics of the basidiocarp. Other characters such as pileus margin, pileus texture, context thickness, and hymenium length show lower loadings, indicating that they contribute less to the separation of specimens in the PCA. Yamamoto et al. (2014) stated that loadings values greater than about 0.2 (≥0.2) indicate that the variable has a meaningful contribution to the component, whereas loadings lower than 0.2 (<0.2) indicate a weak contribution. The traits are highly variable in *S. commune* and reflect the species phenotypic plasticity, even among closely related isolates (Marian et al., 2024). Variation in basidiocarp size is common in *S. commune* because fruiting body development is influenced by environmental conditions, substrate availability, and developmental stages of the fungus. Studies on mushroom-forming fungi have shown that fruit body morphology, including cap size and structural organization, can vary substantially depending on genetic and environmental factors affecting basidiocarp development (Kües & Navarro-González, 2015). In addition, population studies have shown that *S. commune* possesses high genetic diversity worldwide, which may contribute to observable morphological variability among specimens (James et al., 2011).

4. CONCLUSION

S. commune from Java Island, Indonesia, demonstrates a wide geographic distribution and broad host range, with *Swietenia mahagoni*, *Albizia chinensis*, and *Rubus idaeus* reported here as new host, and displayed three pileus shape types orbicular, suborbicular, and predominantly flabelliform which reflecting high morphological plasticity. ITS rDNA barcoding, UPGMA clustering, and PCA analyses consistently showed close relationships among isolates and indicated that pileus morphology and fruit body attachment characteristics were the main contributors to intraspecific variation.

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CONFLICT OF INTEREST
The authors declare no conflicts of interest.

AUTHOR CONTRIBUTION
Kusrinah: Conceptualization, methodology, data analysis, and writing original draft; Budi Setiadi Daryono: Supervision, review and editing; Purnomo: Supervision, review and editing; Rina Sri Kasiamdari: Supervision, conceptualization, writing, final review and editing, and corresponding author.

DATA AVAILABILITY
All data generated or analysed during this study are included in this published article.

DECLARATION OF GENERATIVE AI

During the preparation of this work, the author used ChatGPT in order to assist in drafting and in finding reliable references. After using the tool, the author reviewed and edited the content as needed to improve the sentence and grammar of the manuscript. The authors take full responsibility for the content of the publication.

ETHICS

Not applicable.

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