New Record of Edible Chicken of the Wood Mushroom, *Laetiporus* versisporus (Lloyd) Imazeki (Fomitopsidaceae, Polyporales) from Sabah (Northern Borneo), Malaysia

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ABSTRACT

The genus *Laetiporus* has been previously reported from Mesilau in Sabah, Northern Borneo in 1964. To date, no further documentation of the *Laetiporus* genus has been reported in Sabah, Malaysia. This study provides an overview of recent literature on taxonomic updates, distribution and sequence data of *Laetiporus* in Malaysia. During the period March — June 2020, two *Laetiporus* specimens were collected in Maliau Basin Conservation Area and Sipitang. These two specimens were identified as *L. versisporus* based on morphological characteristics and molecular methods. Interestingly, no sequence data for this particular species have been documented for Malaysian Borneo. This study represents the initial documentation of *L. versisporus* in Sabah (Northern Borneo) that have potential applications in medicine and food industry and provide insights into its phylogenetic relationship within the genus *Laetiporus*.

Keywords: Brown-rot fungi, chicken of the wood, Northern Borneo, wood-decaying fungi

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INTRODUCTION

The genus *Laetiporus* Murrill belongs to the family Fomitopsidaceae and the order Polyporales. *Laetiporus* is a parasitic bracket fungus, it is widely distributed around the world from temperate to tropical region (Murrill, 1904; Sinclair *et al.*, 1987). While some species within this genus are recognised as forest pathogens, others are edible and have medicinal properties (Dai *et al.*, 2007; 2009; Ota *et al.*, 2009; Banik *et al.*, 2012; Song *et al.*, 2014; Elkhateeb *et al.*, 2021; Hassan *et al.*, 2021).

Previous research has identified 15 accepted species of *Laetiporus* globally, and phylogenetic analyses have confirmed the presence of 11 species within L. sulphureus complex. The species are commonly associated with various families, including Betulaceae, plant Elaeocarpaceae, Burseraceae. Fabaceae. Fagaceae, Meliaceae, Myrtaceae, Oleaceae, Pinaceae, Salicaceae, Sapindaceae, Taxaceae (Burdsall & Banik, 2001; Lindner & Banik, 2008; Ota *et al.*, 2009; Vasaitis *et al.*, 2009; Banik *et al.*, 2012; Song *et al.*, 2014). *Laetiporus* spp. have been recognised as brown rot fungi which can cause brown cubical heart rot (Sinclair *et al.*, 1987), thereby playing a role in the cycle of forest ecosystem (Ota *et al.*, 2009; Song *et al.*, 2014).

Laetiporus spp. have a rich culinary tradition and have been consumed as food for many years, especially in North America, Europe and Japan. The species are highly regarded for their culinary, medicinal and ecological importance. Some are recognised as valuable source of essential vitamins and minerals, including vitamins B & D, copper, potassium and selenium. With low calories and fat, this species is a nutritious dietary choice (Luangharn et al., 2014a).

In tropical regions, *Laetiporus* spp. exhibit a range of colours, with their pileus surfaces ranging from white to reddish orange, and their pore surfaces are typically white to pale buff.

Imazeki and Hongo (1989) revised East Asian Laetiporus and found that there are at least three species, L. sulphureus var. sulphureus and L. versisporus and variety, L. sulphureus var. miniatus. Overeem (1925) identified a tropical Asian population with a reddish pileus as L. miniatus, which later became junior synonym with L. sulphureus. On the other hand, L. versisporus (Lloyd) Imazeki displays a different set of morphological characteristics. Initially, the basidiocarps are lemon-yellow to white in color, but they undergo a color change as they mature, turning white to brown. Occasionally, L. versisporus may generate incomplete tubes and basidiospores, which could represent an intermediate form between L. sulphureus and L. versisporus (Ota & Hattori, 2008).

In Sabah (Northern Borneo), Corner (1984) made the initial discovery of the genus *Laetiporus* near Mount Kinabalu in Mesilau, Sabah (Northern Borneo). The publication

provided a detailed account of the genus's morphology and taxonomy, emphasizing its smooth upper surface and white under surface, which produces spores. It should be noted that Corner (1984) reported the collections of mainly L. discolor, L. discolor var. brunnescens, and L. discolor var. pallidus at Mount Kinabalu (Mesilau) (Figure 1). On the other hand, L. persicinus and L. sulphureus were found exclusively in Peninsular Malaysia (Corner, 1984). Later, Yamashita et al. (2018) reported the presence of L. discolor and L. sulphureus in Sarawak (Figure 1), while Lee et al. (2012) documented L. sulphureus in Peninsular Malaysia (mainland). The species occurrences and their edibility information of Laetiporus in Malaysia were not documented in any later studies. The present taxon has been previously reported from China and Japan. However, it has not been reported from Malaysia prior to this study.

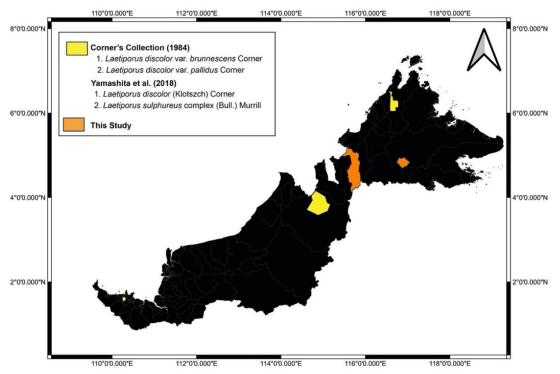


Figure 1. *Laetiporus* collection from Sabah (Northern Borneo) based on Corner's study (Corner, 1984) and Sarawak by Yamashita *et al.* (2018). New collections from recent study are updated (orange color)

The objective of this study was to gather and analyse data on the Bornean *Laetiporus* species recently discovered in the Maliau Basin and Sipitang regions. The investigation is focused on examining the morphological characteristics of these specimens and determining their

phylogenetic relationship with other known taxa found worldwide. Thus, the study will provide valuable insights to the taxonomy and evolutionary relationships of the recently collected Bornean *Laetiporus*.

MATERIALS AND METHODS

Study Sites and Sample Collections

The Laetiporus specimens were collected from a fruit orchard in Sipitang and primary low dipterocarp forest of Maliau Basin Conservation Area (MBCA) in Sabah, between March and June 2020. The sporocarps were photographed in the field, labeled with field number, the morphological characters and information of the substrate were recorded. Key colours were based on the Methuen Handbook of Color (Kornerup & Wanscher, 1978). The specimens were then dried using food dehydrators (Primada Food Dehydrator MPD68) at 40 °C for 24 hours, stored in ziplock bags with anhydrous silica gels. The specimens were then brought to the laboratory for further analysis and curated specimens were deposited at the BORNEENSIS (BORH) herbaria, Institute of Tropical Biology and Conservation (ITBC), Universiti Malaysia Sabah.

The macro-morphological characteristics such as the colour and shape of fruiting body and the colour of the pore bearing surface were recorded based on field notes. Micromorphological data were obtained from dried specimens and observed under a compound microscope. Sections were prepared and mounted with 3% KOH and stained with Meltzer's reagent and Congo Red. The microscopic characters were studied at a magnification up to 1,000 x and photographed using an 80i Nikon (Nikon, Tokyo, Japan). A total of 100 basidiospores, 20 basidia were abbreviations measured. The for measurements (x/y/z) denote 'x' basidiospores measured from 'y' basidiocarps of 'z' specimens. Q is the length/width ratio of basidiospores, is given as $Q_m \pm$ standard deviation where Q_m is the average of all Q basidiospores. Scanning Electron Microscope (SEM) was used for basidiospore images. The voucher specimen examined was preserved and added to the BORNEENSIS (BORH) with the accession number (BORH/F/UMS-00150 and BORH /F/UMS-00151).

Culture of *Laetiporus versisporus*

Pure cultures were made using sterilised inoculation needle from internal tissues of fresh sporocarps into potato dextrose agar (PDA) and

incubated at room temperature 25 °C for 14 days. The mycelial growth was determined by using a ruler across the plate and calculated the average of the vertical and horizontal colony diameter (Luangharn *et al.*, 2014b). Colony morphology was observed for 14 days. The growth rate was calculated by the ratio of colonies diameter and time (days).

DNA Extraction, PCR Amplification and Sequencing

The fungal specimens used in this study are listed (Table 1). Genomic DNA isolation and PCR of the studied material were done at ITBC, Universiti Malaysia Sabah. Genomic DNA (10 – 50 mg) were extracted from dried specimens using E.Z.N.A Fungal DNA Kit (Omega Bio-Tek, USA). The detailed methods were based on the manufacturer's protocol. The internal transcribed spacer regions (ITS) were amplified with fungal primer pair ITS1F/ITS4 (White et al., 1990) and the large subunit of nuclear ribosomal RNA gene (nLSU) was amplified with fungal primer pair LROR/LR7 (Vilgalys & Sun, 1994). Gene regions were amplified in 50 μL reactions containing 3 µL template DNA, 27.75 μL ddH₂0, 4 μL of primers and 15.25 μL PCR mix were performed in a C1000 thermal cycler (Bio-Rad, Hercules, CA). The PCR amplicons were then sent to Apical Scientific (Seri Kembangan, Selangor) for Sanger sequencing.

Phylogenetic Analysis

The newly generated sequences (ITS = 2; LSU = 2) were subjected to Basic Local Alignment Search Tool (BLASTn). All sequences were determined in both directions to produce consensus sequences with BioEdit Sequence Alignment Editor Version 7.2.5 (Hall, 1999). All newly generated sequences were submitted to GenBank (Table 1). A total of 53 sequences derived from GenBank were used for the phylogenetic analyses, and Antrodia serialis (Fr.) Donk and Fomitopsis pinicola (Sw.) P. Karst were used as the outgroup (Song et al., 2018). Sequences were manually edited and aligned using AliView (Larsson, Maximum likelihood (ML) was performed for both gene regions separately by using RAxML-HPC2 version 8.2.12 (Stamatakis, 2006) with GTR model and 1,000 rapid bootstrap replicates. Bayesian inference analysis was run using MrBayes 3.2 (Ronquist et al., 2012) and the best fit model of nucleotide evolution was estimated using JModelTest2 in CIPRESS portal based on AIC (Akaike Information Criterion) and the selected models were GTR+1+G. Bootstrap values (BS) (\geq 70%) and posterior probability (PP) (\geq 90%) were considered significantly

supported. Phylogenetic trees were visualised using FigTree v1.4.4 and the tree was edited using Adobe Illustrator v27.6.1. Sequence alignment was deposited at TREEBASE (http://purl.org/phylo/treebase; submission ID 30528).

Table 1. A list of species, specimens and GenBank accession numbers of sequences used in this study

Species Collection No.		Origin	GenBank Accessions 28S (LSU)	References	
Antrodia serialis	Cui 10519	China	KP715323	Yuan-Yuan & Cui (2016)	
Fomitopsis pinicola	Cui 10405	China	KC844857	Unpublished	
Laetiporus ailaoshanensis	Dai 15624	China	KX354495	Song & Cui (2017)	
L. ailaoshanensis	Dai 13574	China	KX354496	Song & Cui (2017)	
L. ailaoshanensis	Cui 12387	China	KX354497	Song & Cui (2017)	
L. ailaoshanensis	Dai 13567 (Paratype)	China	China KX354498		
L. ailaoshanensis	Dai 13566	China	KX354499	Song et al. (2014)	
L. ailaoshanensis	Dai 13256 (Holotype)	China	KF951317	Song et al. (2014)	
L. cincinnatus	Dai 12811	USA	KF951304	Song et al. (2014)	
L. cincinnatus	JV 0709/168J	USA	KF951305	Song et al. (2014)	
L. conifericola	JV 0709/81J	USA	KF951327	Song et al. (2014)	
L. conifericola	HHB15411	Canada	KX065982	Unpublished	
L. cremeiporus	Cui 10586	China	KF951297	Song et al. (2014)	
L. cremeiporus	Li 140927	China	KX354485	Song & Cui (2017)	
L. cremeiporus	Dai 10107	China	KF951301	Song et al. (2014)	
L. cremeiporus	Cui 10991	China	KF951298	Song et al. (2014)	
L. gilbertsonii	JV 1109/31	USA	KF951306	Song et al. (2014)	
L. gilbertsonii	CA 13	USA	EU402527	Lindner & Banik (2008)	
L. gilbertsonii	TJV 2000/101	USA	EU402528	Lindner & Banik (2008)	
L. gilbertsonii	CA6	USA	KX065984	Unpublished	
L. huroniensis	CBS:136051	USA	MH877606	Vu <i>et al.</i> (2019)	
L. huroniensis	HMC1	USA	KX065985	Unpublished	
L. montanus	Cui 10011	China	KF951315	Song <i>et al.</i> (2014)	
L. montanus	Dai 15888	China	KX354494	Song & Cui (2017)	
L. montanus	Cui 10015	China	KF951311	Song et al. (2014)	
L. montanus	LCB_524	Germany	OK481090	Unpublished	
L. montanus	BRNM:706688	Czech Republic	EU884419	Tomsovsky & Jankovsky (2008)	
Laetiporus sp.	EUC 1	USA	EU402541	Lindner & Banik (2008)	
Laetiporus sp.	KOA 1	USA	EU402542	Lindner & Banik (2008)	
Laetiporus sp.	Cui 12219 (Paratype)	China	KX354500	Song & Cui (2017)	
Laetiporus sp.	Cui 12240 (Holotype)	China	KX354501	Song & Cui (2017)	
Laetiporus sp.	Cui 12390 (Paratype)	China	KX354502	Song & Cui (2017)	
L. sulphureus	Cui 12389	China	KX354487	Unpublished	
L. sulphureus	Cui 12388	China	KX354486	Unpublished	
L. sulphureus	JV 1106/15	Czech Republic	KF951303	Song <i>et al.</i> (2014)	
L. sulphureus	Dai 12154	Czech Republic	KF951302	Song et al. (2014)	
L. sulphureus	Cui 12370	China	KX354504	Song & Cui (2017)	
L. sulphureus	Cui 12371	China	KX354505	Song & Cui (2017)	
L. sulphureus	Z.R.L. CA04	Canada	KX354506	Song & Cui (2017)	
L. sulphureus	Z.R.L. CA04 Z.R.L. CA08	Canada	KX354507	Song & Cui (2017)	
L. sulphureus	TFRI 1092	-	EU232302	Unpublished	
L. sulphureus	DA-41	USA	KC585183	Ortiz-Santana et al. (2013)	
L. versisporus	Cui 7882	China	KF951323	Song et al. (2014)	
L. versisporus	Dai 13160	China	KF951320	Song et al. (2014)	
L. versisporus	Cui 9154	China	KF951322	Song et al. (2014)	
L. versisporus	Cui 5488	China	KF951321	Song et al. (2014)	
L. versisporus	Dai 10992	China	KF951325	Song et al. (2014)	
L. versisporus	Dai 13052	China	KF951324	Song et al. (2014)	
L. versisporus	BORHF0050	Sabah, Malaysia	OQ954850	This study	
L. versisporus	BORHF0051	Sabah, Malaysia	OQ954849	This study This study	
L. xinjiangensis	Dai 15953 (Holotype)	China	KX354488	Song & Cui (2017)	
L. xinjiangensis	Dai 15828 (Paratype)	China	KX354489	Song & Cui (2017)	
L. xinjiangensis	Dai 15898A (Paratype)	China	KX354469 KX354492	Song & Cui (2017) Song & Cui (2017)	
	· • • • • • • • • • • • • • • • • • • •	China		Song & Cui (2017) Song & Cui (2017)	
L. xinjiangensis L. zonatus	Dai 15825 (Paratype) Dai 13633	China	KX354493		
	HKAS 71806 (Paratype)		KX354508	Song & Cui (2017)	
L. zonatus	SAAS 547	China China	KF951310	Song <i>et al.</i> (2014) Song & Cui (2017)	
L. zonatus	SAAS 547 SAAS 681	China	KX354509	Song & Cui (2017) Song & Cui (2017)	
L. zonatus			KX354510	_	
L. zonatus	Cui 10403 (Paratype)	China	KF951307	Song <i>et al.</i> (2014)	

RESULTS

New Distribution Record of *Laetiporus*

The Laetiporus species in Malaysia mostly found on rotting woods in the mountainous region. Laetiporus versisporus occurrence in different location from Sabah has been recorded and mapped with previously known records (Figure 1). Both samples were found at lowland forest (MBCA) and nearby fruit orchard (Sipitang). The two new collections were updated in the Northern Bornean Agaricomycetes project under Borneensis Herbarium. It was noted that one of the collections (BORH/FUMS-00151) was found on a living tree of Artocarpus (Family: Moraceae) whereas the other collection (BORH/FUMS-00150) was from an unidentified dead log found at Maliau Basin primary forest area. Both L. versisporus collections were found below elevation 700 m a.s.l (Table 2).

Taxonomy

Laetiporus versisporus (Lloyd) Imazeki (Figure 2)

Type locality: Miyazaki pref., Miyazaki-gun, Tano-cho, Miyzaki Univ., S. Kurogi, F-19732 (TFM)

Taxon names

- = *Calvatia versispora* Lloyd (1915).
- = Polyporus calvatoides Imazeki (1940).

Ecology and Distribution: Fruiting gregariously on living and dead logs. Known from Central China, South China (Song *et al.*, 2014), tropical and subtropical areas in Japan, Taiwan, South Korea (Ota *et al.*, 2009; Song & Cui, 2017) and Australia (Mushroom observer).

Specimens Examined: Malaysia. Sabah, Tongod, Maliau Basin Conservation Area (MBCA), 4.8531° N, 116.8439° E, 3 March 2020, BORH/FUMS-00150; Sipitang, 5.0792° N, 115.5508° E, 28 June 2020, BORH/FUMS-00151.

Substrate: On deadwood (BORH/FUMS-00150), living *Atrocarpus* sp. (Moraceae) tree (BORH/FUMS-00151).

Description: The basidiocarp of *L. versisporus* is centrally stipitate, occasionally imbricate, single circular pileus or in rosette with multiple pilei up to 23 cm in diameter, upper surface yellow orange (5A6 - 5A7), pore surface white (5A1) yellowish to the margin (5A2), angular pores 0.2 - 0.3 cm, with thin dissepiments (Figure 2). Hyphal system dimitic, contextual generative hyphae thin-walled, hyaline, simpleseptate, with rare branching, 8 - 14 µm in diameter, contextual binding hyphae solid-to thick-walled, hyaline, non-septate, branched and interlocking 18 µm in diameter. Cystidia or other sterile hymenial elements lacking. Basidia clavate, 2 – 4 sterigmata, 13 – $17 \times 3 - 4 \mu m$, simple-septate at the base. Basidiospores ovoid to short ellipsoid, (2.6-)2.7 $-4.8 (-5.0) \times (2.9-) 3.0 - 4.8 (-5.0) \mu m$, Q = 1.11 μm (Figure 2).

Sequence: LSU (OQ954849 (BORH/F00150), OQ954850 (BORH/F00151)); ITS (OQ832657 (BORH/F00150), OQ832658 (BORH/F00151)).

Remarks: Bornean collections have smaller size of basidiospore and basidia, pore surface white and yellowish to the margin. *Laetiporus versisporus* found from this study differ from *L. discolor, L. persicinus*, and *L. sulphureus* in having smaller ovoid to short ellipsoid spores and basidiospores, where other *Laetiporus* species have all distinctly larger spores (Table 2).

Culture Morphology

Both strains of L. versisporus exhibit rapid growth $(6-9 \text{ cm}^{d-1})$ on PDA plates within a span of 14 days, producing a dense, cottony and fluffy mycelial mat that ranges in color from white to yellowish orange (Figure 3). The mycelial mat appeared smooth and fragmented. Crystals typically present, especially in agar, bipyramidal to prismatic.

Phylogenetic Analysis

Phylogenetic tree for each gene (ITS = 2; LSU = 2) was constructed and both genes were congruent. In this study, the LSU dataset was used as the main figure (ITS dataset not shown). The alignments for LSU had an aligned length of 892 characters, of which 791 characters were constant, 85 were parsimony-informative and 87 were parsimony-uninformative. The LSU phylogeny within the related taxa of *Laetiporus*

showed that the two specimens of *Laetiporus* spp. in this study, were closely related to the Chinese *L. versisporus* with high support value (99% BS; 0.99 PP) (Figure 4). The topology of the phylogenetic tree obtained from the Bayesian analysis and the ML analyses were identical with

little variation in the statistical support. Hence, the phylogenetic tree generated using ML has been displayed in Figure 4. Thus, the phylogenetic data together with morphological analysis, showed that the two specimens were closest match to *L. versisporus*.

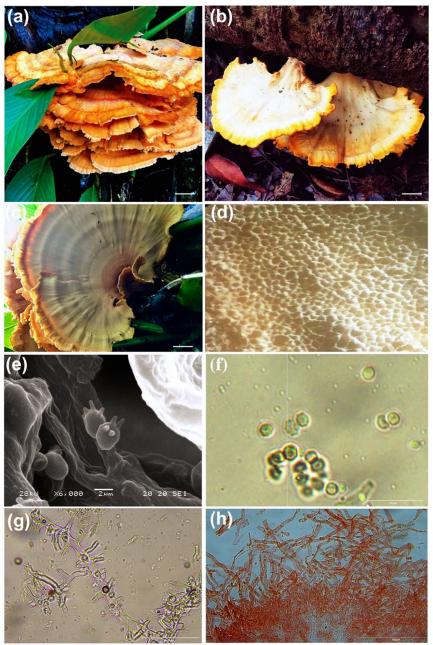


Figure 2. (a – c) Basidiocarps of *Laetiporus versisporus*, (d) Pore surface. Scale bar of a, b, c, d - 1cm; Scale bar of d - 0.5cm (e) Basidia. (f) Basidiospores. (g) Skeletal hyphae (h) Generative hyphae. Scale bar of a, b, c- 1 cm; Scale bar of d- 5 mm; Scale bar of e- 2 μ m; Scale bar f, g, h- 100 μ m

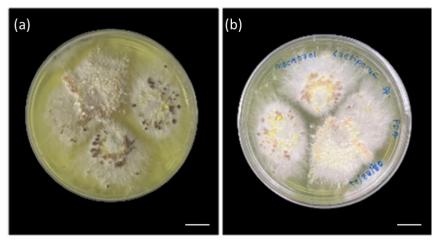


Figure 3. Mycelia growth of *Laetiporus versisporus* strains: (a) BORH/FUMS-00151 (b) strain BORH/FUMS-00150 on media plates (PDA) incubated at room temperature for 14 days. Scale bar of a & b-1 cm

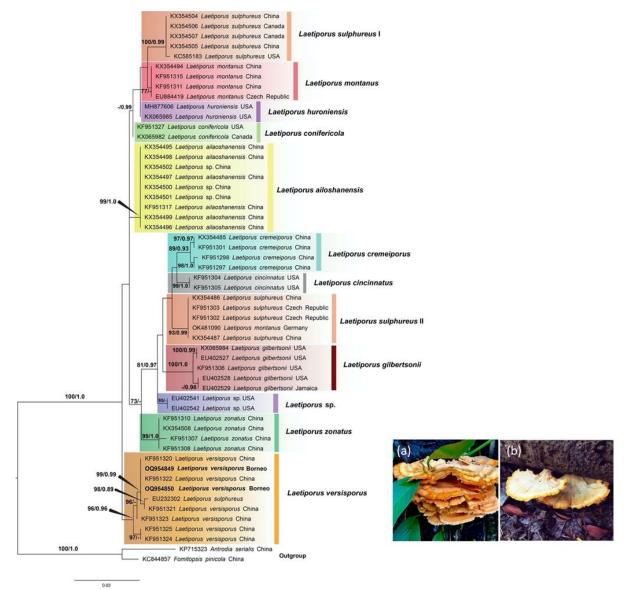


Figure 4. Maximum Likelihood (ML) analysis of LSU of *Laetiporus* spp. and related taxa. Support values are obtained from ML bootstrap values (BS≥70) and Bayesian Posterior Probability (PP≥0.90). Sequences from this study are indicated in bold. Sequences from this study are indicated in bold. The Laetiporus versisporus sample from Sipitang (a) OQ954850 and sample from MBCA (b) OQ954849

Table 2. List of Laetiporus species recorded in Malaysia

Species	Pileal surface	Pore surface	Pores	Basidiospores	Substrate/Host	Location	References
Laetiporus discolor (Klotzsch) Corner	NA	NA	NA	NA	NA	Sarawak – Mt Serapi	Yamashita et al. (2018)
L. discolor var brunnescens Corner	White, alutaceous to pallid ochraceous	Ochraceous when fresh	4-7 mm	Ovoid to ellipsoid 4.0 -6.3 x 2.8-4.5 μm	On rotting logs in the forest	Sabah-Mt Kinabalu	Corner (1984)
L. discolor var pallidus Corner	White, alutaceous to pallid ochraceous with pale pinkish yellow to pale orange zone	White, alutaceous with pale pinkish or orange zone	4-7 mm	Ovoid to ellipsoid 4.0-5.5 x 3.0 -4.5 μm	On rotting trunks in the forest	Sabah-Mt Kinabalu, Mesilau, Bembangan Valleys.	Corner (1984)
L, persicinus	Brown to pinkish velvety texture	White and bruises when touch	2-4 mm	Ovoid to ellipsoid	On the ground in gardens, plantations and secondary forest	Peninsular Malaysia; Pahang-Tembeling	Corner (1984)
L. sulphureus (Bull.) Murrill	Bright orange	Bright creamy yellow	2-4 mm	Ovoid to ellipsoid 5.0-6.8 x 4.0-5.0 µm	On rotting trunks in the forest	Pahang; Perak	Lee et al. (2009)
L. sulphureus (Bull.) Murrill	NA	NA	NA	NA	NA	Sarawak – Mt Mulu; Apat Camp River	Yamashita et al. (2018)
Laetiporus versisporus (BORH/F00150)	Yellow orange	White, yellowish to the margin	2-3 mm	Ovoid to short ellipsoid 2.6-5.0 x 2.9-5.0 μm	On decaying log	Sabah – Maliau Basin Conservation Area (MBCA)	This study
Laetiporus versisporus (BORH/F00151)	Yellow orange	White, yellowish to the margin	2-3 mm	Ovoid to short ellipsoid $2.6-5.0 \times 2.9-5.0 \mu m$	On Artocarpus tree.	Sabah-Sipitang	This study

DISCUSSION

study utilised a combination of morphological and molecular methods identify various Laetiporus species found in Sabah (Northern Borneo). Distinguishing between species within the Laetiporus genus can be challenging solely based on morphological characteristics such as the color, shape, and size, as different species can exhibit variations in traits. Molecular techniques employed to aid in the species differentiation to provide a more accurate identification of the species. The morphological characteristics of the Laetiporus species found in Sabah (Northern Borneo) are consistent with those of *Laetiporus* versisporus described by Imazeki (1943) from Japan, which typically has a yellow to orangecolored basidiocarps that is centrally stipitate and can be either single circular pileus or in rosette form with multiple pilei. However, the Bornean L. versisporus differs from the L. versisporus from China and Japan in having smaller basidiospores $(2.6 - 5.0 \times 2.9 - 5.0 \mu m)$. To date, only three *Laetiporus* species have been reported in Malaysia, Laetiporus discolor, Laetiporus persicinus and Laetiporus sulphureus (all without molecular data). This study presents new record of L. versisporus in Sabah (Northern Borneo), where this species mostly found in the East Asia from the Yunnan-Guizhou Plateau, Hanan to Japan and South Korea (Ota et al., 2009; Banik et al., 2012). There is limited information available on L. versisporus compared to other species of Laetiporus. The known substrates of L.

versisporus are dead logs, stumps and living trees (*Castanopsis* and *Quercus*) (Ota *et al.*, 2009). These findings adds new information on the host tree and the association of *L. versisporus* with a fruit tree (*Artocarpus*).

versisporus shares similar Laetiporus morphological characteristics with sulphureus, as both have orange-yellowish caps and white-yellowish pore surfaces. However, L. sulphureus produces larger basidiospores (5.0 - $6.8 \times 4.0 - 5.0 \ \mu m$) and has larger pores (2 – 4 mm). The morphological characteristics of the other Malaysian (Peninsular) *Laetiporus* species are consistent with the descriptions provided by Ota et al. (2009). Detailed morphological descriptions of the species reported by Yamashita et al. (2018) are not available. Nonetheless, this genus exhibits certain shared morphological characteristics and similarities (Song et al., 2014). Therefore, molecular identification is needed to confirm the species.

In this study, we determined the Bornean *L. versisporus* were closely related to *L. versisporus* from China with a high level of support (99% BS/99% PP) even though there is a slight difference in the morphological characteristics. This species can be found in Eastern Asia ranging from cool temperate to subtropical areas including Japan, Korea (Vasaitis *et al.*, 2009) and China (Hattori & Zang, 1995; Dai *et al.*, 2007). This study confirms that *L. versisporus* is recognised as a distinct species among the various taxa of *Laetiporus* (Ota *et al.*, 2009).

The L. sulphureus complex in East Asia represents a group of species that are morphologically and ecologically distinct (Ota et al., 2009, Song et al., 2014). This study confirms that L. sulphureus remains paraphyletic, consisting of two different lineages. The L. sulphureus lineage I occur on both hardwoods and conifers and L. sulphureus II restricted to hardwoods (Vasaitis et al., 2009). However, the complexity of the L. sulphureus complex remains unresolved and further investigation is needed. Further sampling work on Laetiporus sensu stricto in Peninsular Malaysia and Malaysian Borneo is needed. In addition, molecular data should be incorporated to verify the species level identification.

Siwulski *et al.* (2009) reported that PDA was found to be the optimal agar medium for mycelium growth of *L. sulphureus*. In this study, we successfully grew the pure culture of *L. versisporus*. Both strains grew rapidly and colonized the PDA media in 14 days. The mycelial growth was dense and fluffy for both collections. Growing *Laetiporus* on culture plates is an essential tool for studying and utilizing the fungus and it also can provide valuable insights into its biology, ecology and potential applications for domestication of the Bornean *Laetiporus*.

Laetiporus fungi have been the subject of several studies that have contributed valuable knowledge on their taxonomy, ecology, genetic diversity and medical properties. These fungi have shown potential as sources of bioactive compounds, including anti-inflammatory, antitumor and immunomodulatory compounds, making them promising candidates for medicinal applications (Duan et al., 2022). Researchers have identified various compounds Laetiporus species, such as polysaccharides, terpenoids and phenolic compounds, which have potential applications in medicine (Petrović et al., 2014). Further research is needed to analyze the chemical composition and nutritional properties of the Borneo L. versisporus, which could provide valuable information on their potential uses.

CONCLUSION

This study provides information on the new record of "chicken-of-the-woods" mushroom in Borneo. The mushrooms were identified as Laetiporus versisporus based on its morphology and phylogenetic analysis of ML and Bayesian PP. The phylogenetic tree of LSU within the Laetiporus species have shown that the Bornean Laetiporus (BORH/F00150; BORH/F00151) were closely related to the *L. versisporus* from China with strong support values (99% BS / 99% PP).

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REFERENCES

- Banik, M., Lindner, D., Ortiz-Santana, B. & Lodge, D. (2012). A new species of *Laetiporus* (Basidiomycota, Polyporales) from the Caribbean basin. *Kurtziana*, 37: 15-21.
- Burdsall, H.H. & Banik, M.T. (2001). The genus *Laetiporus* in North America. *Harvard Papers in Botany*. 6: 43-55.
- Corner, E.J.H. (1984). Ad Polyporaceas III. Piptoporus, Buglossoporus, Laetiporus, Meripilus and Bondarzewia. *Beihefte zur Nova Hedwigia*, 78: 133-222.
- Dai, Y.C., Cui, B.K., Yuan, H.S. & Li, B.D. (2007). Pathogenic wood-decaying fungi in China. *Forest Pathology*, 37: 105-120. DOI: 10.1111/j.1439-0329.2007.00485.x
- Dai, Y.C., Yang, Z.L., Cui, B.K., Yu, C.J. & Zhou, L.W. (2009). Species diversity and utilization of medicinal mushrooms and fungi in China (Review). *International Journal of Medicinal Mushrooms*, 11(3): 287-302. DOI:10.1615 /intjmedmushr.v11.i3.80
- Duan, Y., Qi, J., Gao, J.M. & Liu, C. (2022). Bioactive components of Laetiporus species and their pharmacological effects. *Applied Microbiology and Biotechnology*, 106: 5929-5944. DOI: 10.1007/s00253-022-12149-w

- Elkhateeb, W., Elghwas, D., Gundoju, N., Tiruveedhula, S., Akram, M. & Daba, G. (2021). Chicken of the woods *Laetiporus sulphureus* and *Schizophyllum* Commune treasure of medicinal mushrooms. *Journal of Microbiology and Biotechnology*, 6: 1-7. DOI: 10.23880/oajmb-16000201
- Hall, T.A. (1999). BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series*, 41: 95-98.
- Hassan, K., Matio Kemkuignou, B. & Stadler, M. (2021). Two new triterpenes from Basidiomata of the medicinal and edible mushroom, *Laetiporus sulphureus*. *Molecules*, 26(23): 7090. DOI: 10.3390/molecules26237090
- Hattori, T. & Zang, M. (1995). List of polypores collected in East China. *Bulletin of the National Museum of Nature and Science, Series B (Botany)*, 21: 95-105.
- Imazeki, R. (1940). Observations on Japanese fungi IV. *Journal of Japanese Botany*, 16: 264-272.
- Imazeki, R. (1943). Genera of Polyporaceae of Nippon. *Bulletin of Tokyo Science Museum*, 6: 1-111.
- Imazeki, R. & Hongo, T. (1989). Colored illustrations of mushrooms of Japan Vol. II. Hoikusha, Osaka.
- Kornerup, A. & Wanscher, J.H. (1978). *Methuen handbook of colour*. Eyre Methuen.
- Larsson, A. (2014). AliView: A fast and lightweight alignment viewer and editor for large datasets. Bioinformatics.
- Lee, S.S., Alias, S.A., Jones, E.G.B., Zainuddin, N. & Chan, H.T. (2012). Checklist of fungi of Malaysia, Forest Research Institute Malaysia (FRIM), Malaysia. Perpustakan Negara Malaysia. pp. 556.
- Lee, S., Chang, Y. & Noraswati, M. (2009). Utilization of macrofungi by some indigenous communities for food and medicine in Peninsular Malaysia. *Forest Ecology and Management*, 257: 2062-2065. DOI: 10.1016/j.foreco.2008.09.044
- Lindner, D.L. & Banik, M.T. (2008). Molecular phylogeny of *Laetiporus* and other brown-rot polypore genera in North America. *Mycologia*, 100: 417-430. DOI: 10.3852/07-124r2
- Luangharn, T., Hyde, K.D. & Chukeatirote, E. (2014a). Proximate analysis and mineral content of *Laetiporus sulphureus* strain MFLUCC 12-

- 0546 from Northern Thailand. *Chiang Mai Journal Science*, 41(4): 765-770. DOI: 10.1080/21501203.2014.957361
- Luangharn, T., Karunarathna, S.C., Hyde, K.D. & Chukeatirote, E. (2014b). Optimal conditions of mycelia growth of *Laetiporus sulphureus* sensu lato. *Mycology*, 5(4): 221-227. DOI: 10.1080/21501203.2014.957361
- Murrill, W.A. (1904). The Polyporaceae of North America IX. Inonotus, Sesia and monotypic genera. *Bulletin of the Torrey Botanical Club*, 31: 593-610.
- Ortiz-Santana, B., Lidner, D.L., Miettinen, O., Justo, A. & Hibbet, D. (2013). A phylogenetic overview of the Antrodia clade (Basidiomycota, Polyporales). *Mycologia*, 105: 1391-1411. DOI: 10.3852/13-051
- Ota, Y. & Hattori, T. (2008). Relationships among three Japanese *Laetiporus* taxa based on phylogenetic analysis and incompatibility tests. *Mycoscience*, 49: 168-177. DOI: 10.1007/s10267-007-0403-3
- Ota, Y., Hattori, T., Banik, M.T., Hagedorn, G., Sotome, K., Tokuda, S. & Abe, Y. (2009). The genus *Laetiporus* (Basidiomycota, Polyporales) in East Asia. *Mycological Research*, 113(11): 1283-300. DOI: 10.1016/j.mycres.2009.08.014
- Overeem, C. (1925). *Icones Fungorum Malayensium*. *Heft XII Polyporaceae*. Im Selbstverlag des Mykologischen Museums in Weesp, Wien.
- Petrovic, J., Papandreou, M., Glamoclija, J., Ciric, A., Baskakis, C., Proestos, C., Lamari, F., Zoumpoulakis, P. & Sokovic, M. (2014).
 Different extraction methodologies and their influence on the bioactivity of the wild edible mushroom *Laetiporus sulphureus* (Bull.) Murrill. Food and Function, 5(11): 2948-2960. DOI: 10.1039/c4fo00727a
- Ronquist, F., Teslenko, M., van der Mark P., Ayres D.L., Darling, A., Höhna, S., Larget, B., Liu, L., Suchard, M.A. & Huelsenbeck, J.P. (2012).
 MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Systematic Biology, 61(3): 539-542.
- Sinclair, W.A., Lyon, H.H. & Johnson, W.T. (1987). *Diseases of trees and shrubs*. Cornell University Press, pp. 575.
- Siwulski, M., Pleszczynska, M., Wiater, A., Wong, J.J. & Szczodrak, J. (2009). The influence of different media on the *Laetiporus sulphureus*

- (Bull.: Fr.) Murr. mycelium growth. *Herba Polonica*, 255: 278-284.
- Song, J., Chen, Y.Y., Cui, B.K., Liu, H.G. & Wang, Y.Z. (2014). Morphological and molecular evidence for two new species of *Laetiporus* (Basidiomycota, Polyporales) from southwestern China. *Mycologia*, 106(5): 1039-1050. DOI: 10.3852/13-402
- Song, J. & Cui, B.K. (2017). Phylogeny, divergence time and historical biogeography of *Laetiporus* (Basidiomycota, Polyporales). *BMC Evolutionary Biology*, 17(1): 102. DOI: 10.1186/s12862-017-0948-5
- Song, J., Sun, Y.F., Ji, X., Dai, Y.C. & Cui, B.K. (2018). Phylogeny and taxonomy of *Laetiporus* (Basidiomycota, Polyporales) with descriptions of two new species from western China. *MycoKeys*, 37: 57-71. DOI: 10.3897/mycokeys.37.26016
- Stamatakis, A. (2006). RAxML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. *Bioinformatics*, 22: 2688-2690. DOI: 10.1093/bioinformatics/btl446
- Tomsovský, M. & Jankovský, L. (2008). Validation and typification of *Laetiporus montanus*. *Mycotaxon*, 106: 289-295.
- Vasaitis, R., Menkis, A., Lim, Y.W., Seok, S., Tomsovský, M., Jankovský, L., Lygis, V., Slippers, B. & Stenlid, J. (2009). Genetic variation and relationships in *Laetiporus sulphureus* s. lat., as determined by ITS rDNA sequences and *in vitro* growth rate. *Mycological Research*, 113: 326-336. DOI: 10.1016/j.mycres.2008.11.009

- Vilgalys, R. & Sun, B.L. (1994). Ancient and recent patterns of geographic speciation in the oyster mushroom Pleurotus revealed by phylogenetic analysis of ribosomal DNA sequences. Proceedings of the National Academy of Sciences of the United States of America, 91(10): 4599-4603
- Vu, D., Groenewald, M., de Vries, M., Gehrmann, T., Stielow, B., Eberhardt, U., Al-Hatmi, A., Groenewald, J.Z., Cardinali, G., Houbraken, J., Boekhout, T., Crous, P.W., Robert, V. & Verkley, G.J.M. (2019). Large-scale generation and analysis of filamentous fungal DNA barcodes boosts coverage for kingdom fungi and reveals thresholds for fungal species and higher taxon delimitation. *Studies in Mycology*, 92: 135-154, DOI: 10.1016/j.simyco.2018.05.001
- White, T.J., Bruns, T.D., Lee, S. & Taylor, J. (1990).
 Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In Innis, M.A., Gelfand, D.H., Sninsky, J.J. & White, T.J. (eds.) PCR protocols, a guide to methods and applications. *California Academic Press*: San Diego, 315-322.
- Yamashita, S., Salleh, H. & Hattori, T. (2018). List of polypores collected from natural forests in Sarawak, Borneo Island between 1954 and 2003. *Contributions from the Biological Laboratory Kyoto University*, 31(1): 1-46.
- Yuan-Yuan, C. & Cui, B.K. (2016). Phylogenetic analysis and taxonomy of the *Antrodia heteromorpha* complex in China, *Mycoscience*, 57(1): 1-10. DOI: 10.1016/j.myc.2015.07.003