

Taxonomic Study of the Genus *Marasmiellus* Murrill in Java and Bali

Atik Retnowati, author

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Abstrak

ABSTRACT

Marasmiellus is a tropical and subtropical genus which consists of more than 400 species. The genus plays important roles in ecosystem as a decomposer, and not many information on its economic value. Several monographs of the genus have been published based on morphological data from several tropical forests (neotropics, Africa, Srilanka), and no report on the Indonesian *Marasmiellus*. At present, taxonomic problem occurs in the genus related to the position of *Marasmiellus juniperinus* as a type genus in the phylogenetic tree based on Internal Transcribed Spacer (ITS) region. *Marasmiellus juniperinus* nested into different clade from other *Marasmiellus* species, and it was clustered in *Gymnopus* clade. Since *Marasmiellus* has never been reported from Indonesia and the taxonomic problem occurs at the genus, thus the objectives of this study are 1) To describe the species of *Marasmiellus* found in Java and Bali; 2) To provide an identification key to sections and species; 3) To analyse the phylogenetic relationship within the genus *Marasmiellus* in Java and Bali based on morphological and molecular characters performed by Maximum Parsimony method; 4) To clarify the relationship of the genus *Marasmiellus* and *M. juniperinus* to its closely related genera with inclusion of more *Marasmiellus* spp. based on ITS region of rDNA sequence data; and 5) To describe novel species of *Marasmiellus* based on morphological and molecular data. Java and Bali were chosen as research sites in this study due to several reasons. First, historically most of Indonesian reported agarics were collected from Java, particularly from Mount Gede-Pangrango, Cibodas and Bogor Botanical Gardens. Second, forest degradation is going rapidly in Java and Bali. As a result Indonesia is losing habitats of *Marasmiellus*. This study was divided into three topics according to the purposes of this study. The first topic entitled Species of *Marasmiellus* in Java and Bali. The study was carried out in Herbarium Bogoriense, The Botany Division, Research Center for Biology, The Indonesian Institute of Sciences (LIPI). Herbarium materials are kept in Herbarium Bogoriense (BO) and Harry D. Thiers Herbarium, San Francisco State University (SFSU), SF, CA, USA.

Materials used in this research were collected in 1998-2010 from 10 different locations in Java and Bali which consisted of 22 sites. A representative material of *Marasmiellus juniperinus*, as a type genus, was borrowed from the Herbarium of the University of Tennessee, Knoxville, Tennessee, USA (TENN). The identification results revealed that there were 35 known species; one species as a new combination; 17 undescribed species. Those 35 species consist of 25 species of *Marasmiellus* found in Java, 7 species found in Bali, and 3 species found both in Java and Bali. All described species were treated based on infrageneric classification of Singer (1973) which divided the genus into 10 sections based on morphological characters.

Based on Singer's infrageneric classification *Marasmiellus* in Java and Bali belonged to 5 sections, i.e. sect. *Marasmiellus*, sect. *Rameales*, sect. *Dealbati*, sect. *Candidi*, and sect. *Stenophylloides*. The second topic

entitled Phylogenetic study of the genus *Marasmiellus* based on morphological and molecular analysis. Thirty five morphological characters of 37 taxa were scored manually for phylogenetic analysis based on morphological characters. Those taxa consisted of 35 species of *Marasmiellus* in Java and Bali found (topic 1), a type species of *Marasmiellus* (*M. juniperinus*), and *Crinipellis brunneipurpurea* as an outgroup. Matrix data was analyzed by Phylogenetic Analysis Using Parsimony (PAUP) program. The topology of phylogenetic tree based on morphological data was compared to topology of phylogenetic tree based on molecular data. The phylogenetic tree based on morphological and molecular characters showed that *Marasmiellus* spp. were divided into four major clades. Each clade consisted of several Singer's sections. This result indicated that the sections within Singer's infrageneric classification were polyphyletic. Morphological and molecular phylogenies resulted in this study did not support traditional classification, i.e. Singer's classification of *Marasmiellus*. The phylogenetic relationship of *Marasmiellus* taxa, *Gymnopus* and its closely related genera were analyzed using molecular data of ITS rDNA. Forty one sequences based on ITS region representing of 36 species of *Marasmiellus* were used for phylogenetic analysis. Nine sequences of *Marasmiellus* were generated from this study, and 32 sequences were obtained from the international DNA database (GenBank). The phylogenetic tree showed that *Marasmiellus* taxa were divided into 4 clades. The taxa of *Marasmiellus* and *Gymnopus* were clustered together in three of four clades. The type species, *Marasmiellus juniperinus* was located within the same clade as *Gymnopus fusipes*.

This result indicated that the genus *Marasmiellus* was polyphyletic. The third topic entitled Description of a novel species, *Marasmiellus javanicus*, based on morphological and molecular analysis. *Marasmiellus javanicus* is one of the 17 new species in this study, and it was described by using the integrated morphological and molecular data. *Marasmiellus javanicus* was described as a new species based on 5 specimens found in Bogor Botanical Garden, West Java. Morphologically it was similar to *M. hondurensis* (Murrill) Singer which was collected from Belice in 1906 by Peck (Singer 1973). The characters similarities of *M. javanicus* and *M. hondurensis* were off-white, same size of width, convex with flattened disc of pileus; subdistant lamellae; tomentose and insititious stipe; cutis with diverticulate of pileipellis, and presence of stipe vesture. *Marasmiellus hondurensis* was different from *M. javanicus* in having longer stipe (6-22 mm diam), fusoid basidiospores, Rotalis-type of cheilocystidia, and *M. hondurensis* was found on dicotyledon plant. At present, there is no sequence based on ITS rDNA of *M. hondurensis*. Thus, genetically *M. hondurensis* was unable to be compared to *M. javanicus*. To know the closest species of *M. javanicus*, phylogenetic tree of *Marasmiellus* was constructed. Phylogenetic tree showed that the most closely related species to *M. javanicus* was *M. mesosporus*. *Marasmiellus javanicus* and *M. mesosporus* have ITS rDNA 94% sequence similarity. They differ in 27 nucleotides which were 7 insertion and deletion, and 24 of substitution (A↔G=11, C↔T=13). This study showed that integrated morphological and molecular data are required to identify the *Marasmiellus*.