Development of two novel specific SCAR markers by cloning improved RAPD fragments from the medicinal mushroom *Ganoderma lucidium* (Leysser: Fr) Karst

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ABSTRACT. Development of sequence-characterized amplified region (SCAR) markers from random-amplified polymorphic DNA (RAPD) fragments is a valuable molecular approach for the genetic identification of different species. By using SCAR markers, molecular analysis is reduced to a simple polymerase chain reaction (PCR) analysis using primers designed from the amplicon sequence of RAPD. In this study, the DNA fragments from an improved RAPD amplification of *Ganoderma* species were cloned into a pGM-T vector; positive clones were identified by PCR amplification and enzymatic digestion, and

finally, DNA fragments were sequenced using the Sanger sequencing method for developing the SCAR markers. Two SCAR markers, named LZ4-1 with 534 nucleotides, and LZ5-2 with 337 nucleotides were identified, which are specific to *Ganoderma lucidium* (Leysser: Fr) Karst species. BLAST of these two nucleotide sequences in the GenBank database showed no identity to other species. We deposited these sequences into the GenBank database (LZ4-1 accession No. KM391933, LZ5-2 accession No. KM391934). PCR assays confirmed them as novel molecular markers for *G. lucidium* (Leysser: Fr) Karst, which might be used for genetic authentication of adulterant samples. Thus, our study developed two specific SCAR markers for identifying and distinguishing the medicinal mushroom *G. lucidium* (Leysser: Fr) Karst from other *Ganoderma* species.

Key words: Molecular markers; *Ganoderma lucidum*; Adulterant; Random-amplified polymorphic DNA; Species specific-authentication; Sequence-characterized amplified region