Genome-based Proteomic Analysis of *Lignosus rhinocerotis* (Cooke) Ryvarden Sclerotium

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Abstract

*Lignosus rhinocerotis* (Cooke) Ryvarden (Polyporales, Basidiomycota), also known as the tiger milk mushroom, has received much interest in recent years owing to its wide-range ethnobotanical uses and the recent success in its domestication. The sclerotium is the part with medicinal value. Using two-dimensional gel electrophoresis coupled with mass spectrometry analysis, a total of 16 non-redundant, major proteins were identified with high confidence level in *L. rhinocerotis* sclerotium based on its genome as custom mapping database. Some of these proteins, such as the putative lectins, immunomodulatory proteins, superoxide dismutase, and aegerolysin may have pharmacological potential, while others are involved in nutrient mobilization and the protective antioxidant mechanism in the sclerotium. The findings from this study provide a molecular basis for future research on potential pharmacologically active proteins of *L. rhinocerotis*.

Key words: *Lignosus rhinocerotis*, proteomic analysis, LC-MS, MALDI-MS, proteins.

Introduction

*Lignosus rhinocerotis* (Cooke) Ryvarden (Polyporales, Basidiomycota) is a white-rot fungus that is characterized by having a centrally stipitate pilei arising from the underground tuber-like sclerotium. It is mainly distributed in China, Malaysia, Sri Lanka, the Philippines, Australia, and East Africa [1]; and more commonly known as tiger milk mushroom in Malaysia. In recent years, this mushroom has received much attention owing to its wide-range ethnobotanical uses as a folk medicine. This is also made possible due to the recent success in its domestication of this once very rare and expensive mushroom [2, 3]. This mushroom has been used by the local communities to treat numerous ailments including fever, whooping cough, asthma, cancer, food poisoning, wounds, chronic hepatitis, and gastric ulcers [4, 5].

On-going scientific research has further validated some of the traditional claims on *L. rhinocerotis*. Its petroleum ether, chloroform, methanol, and water sclerotial extracts displayed strong antimicrobial activity against selected human pathogens including gram-positive and gram-negative bacteria and fungi in disk diffusion test [6]. It has also been reported that the aqueous extract of *L. rhinocerotis* sclerotium enhanced neurite outgrowth in PC-12 Adh pheochromocytoma and Neuro-2a mouse neuroblastoma cell lines [7, 8]. Several authors also demonstrated the presence of antiproliferative activity in aqueous (hot and cold) or methanol pressurized liquid extracts, and hot water-soluble polysaccharides isolated from *L. rhinocerotis* sclerotium against human breast carcinoma (MCF7), lung carcinoma (A549) and colorectal cancer (HCT 116) cells, as well as various types of leukemic cells including acute promyelocytic leukemia cells (HL-60), chronic myelogenous leukemia cells (K562), and human acute mononcytoid leukemia cells (THP-1), through apoptosis and/or cell cycle arrest [9-11]. Wong et al. demonstrated that *Polyporus rhinocerus* (synonym to *L. rhinocerotis*) sclerotial polysaccharides exhibited immunomodulatory effects by activation of innate immune cells and T-helper cells in normal and athymic BALB/c mice [12].

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