

Mini Review

The Fascinating Bird's Nest Mushroom, Secondary Metabolites and Biological Activities

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ABSTRACT:

Background: Mushrooms are generous source of nutritional and medicinal compounds. Bird's nest fungi are a gasteromyceteous group of mushrooms named for their similarity in shape to small bird's nests. They are considered from the tiniest and most interesting mushrooms all over the world. It is usually found in shady moist environments, and typically survive on plant debris, soil, decaying wood, or animal's excrement. Bird's nest mushrooms are inedible, though they were not previously reported to be poisonous, due to their tiny size.

Object: this review aims to put bird's nest mushrooms under light spot through describing their morphology and ecology especially of the most common fungus, *Cyathus halleri*. Moreover, discussing important secondary metabolites and biological activities exerted by bird's nest mushrooms.

Conclusion: bird's nest mushrooms are able to produce many novel and potent secondary metabolites that exerted different bioactivities especially as antimicrobial, antitumor, and anti-neuro inflammation activities. Further studies and investigations are encouraged in order to find more about this interesting tiny mushroom.

Keywords: Bird's nest fungi, *Cyathus* spp., secondary metabolites, biological activities.

1. INTRODUCTION

Mushrooms are abundant natural resources that have structurally unique compounds with diverse bioactivities [1-4]. Mushrooms are generally basidiomycetous fungi except for some species. Mushrooms have been reported to be from the most valuable microbes to humankind [5, 6]. Investigations on the therapeutic and nutritional properties of mushrooms are ongoing throughout the world. Researchers are providing vital data on the collection of biologically active secondary metabolites originated from mushrooms. Generally, mushrooms grow wild in many environments around the world and are also commercially cultivated for pharmaceutical and nutritional purposes. Nutritionally, mushrooms are healthy food which is rich in nutrients and vitamins. On the other hand, mushrooms have pharmaceutical and medical applications from centuries especially in Asian countries [7-9].

Recently, isolation and purification of biologically active secondary metabolites from mushrooms has been conducted mainly on the products of fruiting bodies. However, collection of fruiting bodies is a difficult work and is limited

by collecting season and area. Also the small amounts of the collected samples are limited to find metabolites from mushrooms. Thus, mycelial culture of basidiomycetes and ascomycetes originated from wild mushrooms would be suggested as an alternative, dependable, and controllable method that can obtain various bioactive metabolites from mushrooms [10-14].

The genus *Cyathus* Haller ecology and description

Bird's nest fungi are a group of mushrooms named for their similarity to small bird's nests. These fungi can be found worldwide. Bird's nest fungi are saprophytes (microorganisms that live on dead organic matter) [15]. In temperate areas, where conditions are damp and shady, bird's nest fungi can be found anytime of the year. However, autumn is the season in which they are most commonly seen. These tiny fungi are usually identified by the formation of a brownish, greyish, or sometimes whitish outer "nest", carrying brown or white eggs like structures inside it. These eggs like structures are the peridioles which are actually spore-containing structures that act as protective sacs for the mushroom's spores, and are located within the cup-shaped nest-like fruiting body.

Bird's nest fungi are gasteromyceteous fungi, and the most common genera of bird's nest fungi are *Crucibulum* spp., *Cyathus* spp., *Mycocalia* spp., *Nidularia* spp., and *Nidula* spp. These fungi are often found in shaded, moist environments, and typically survive on plant debris, soil, decaying wood, or animals (cows and horses) excrement [15-19].

The characteristic cup and eggs structure (reproductive structure) of the bird's nest fungi is almost metallic looking, like shiny river stones. It provides a unique spores dispersing mechanism. In a storm, the "eggs" are splashed out of the "cups" by raindrops. "Eggs" can travel for a distance as long as about one meter or more before sticking to another object. When the "egg" became dry, it releases fungal spores. When the spores germinate, they grow into branching filaments called hyphae. The mass of hyphae (called mycelium) weaves through moist woody debris and consumes the wood to fuel its growth [16-21].

The genus *Cyathus* Haller (belongs to Division: Basidiomycota; Class: Basidiomycetes; order Nidulariales; Family: Nidulariaceae) can be characterized by its peridium vase-like or infundibuliform to inverted bell-shaped with 3 layers, surface plicate or smooth, often covered with shaggy or tomentose hairs on the outside; peridiole (gleba) lens-shaped and attached to the peridium with a thread-like cord (funiculus); basidiospores hyaline, smooth and mostly with a thick wall, while the shape and size vary [20-23]. In other similar genera like *Nidulaspp.*, and *Crucibulum* spp., *Nidularia* spp., and *Mycocalia* spp. *Nidula* funicular cord with the peridiole is absent which is different than *Cyathus*. Moreover, *Crucibulum* has a single-layered peridium and white to yellowish peridioles, while peridioles appear black to dark brown in *Cyathus* [24-26]. *Cyathus* is the genus with the most species of bird's nest fungi and is represented by 45 species worldwide [27], 14 of which are from India [25-28]. Because bird's nest fungi are harmless to surrounding living plants, animals, or humans, no need to control growth of these mushrooms. Bird's nest fungi are inedible, though they were not previously reported to be poisonous, due to their tiny size. Bird's nest fungi have natural composting abilities and as aforementioned, it isn't harmful to humans, animals or living plants (Figure, 1).



Fig 1: Different *Cyathus* spp. (Bird's nest fungi), Collected by Christine Braaten Locality: United States, Tennessee, Vonore, Kahite. Hosted by <http://mycoportal.org>.

2. BIRD'S NEST FUNGI AS SOURCE OF BIOLOGICALLY ACTIVE COMPOUNDS

Screening of fungi for antimicrobial substances, antitumor substances and others in particular has been greatly enhanced. Ascomycetes and Basidiomycetes mushrooms are known to produce wide range of secondary metabolites. Many interesting compounds were isolated and purified from different species of the basidiomycetous genus, *Cyathus* such as cyathane diterpenoids with unusual 5/6/7 tricyclic skeleton, including their xyloides [29].

The anti-inflammatory, antimicrobial and antitumor compounds isolated from these mushrooms belong to different chemical classes including steroids, terpenes, polysaccharides, fatty acids, phenolic acids, and other metabolites. Taofiq et al., [31], reported that terpenoids, phenolic compounds, and polysaccharides were the most potent compounds causing the anti-inflammatory action exerted by some mushrooms as described in different studies. Diterpenes have been proven to display a wide range of biological properties, including anti-inflammatory [31, 32], antimicrobial [33] antitumor [34]. Previous chemical studies on *Cyathus striatus* resulted in the discovery of antitumor cyathane-type metabolites such as the striatins A, B, C, and D [33, 34].

Liu and Zhang, [35], studied twelve different *Cyathus* species for their capabilities to produce antimicrobial secondary exo-metabolites. These exo-metabolites were able to inhibit some human pathogenic fungi and yeast such as *Aspergillus fumigatus*, *Cryptococcus neoformans*, and *Candida albicans*. The antimicrobial activities of those twelve *Cyathus* species extracts against human pathogenic bacteria were also reported. In another study, the antimicrobial compounds striatins A, B, C were isolated from *Cyathus striatus* during mycelial growth. Striatins A, B, C were highly potent when tested against different Gram positive and negative bacteria, as well as against some fungi imperfecti [36].

Kang et al., [3], reported that three new polyketides, cyathuscavins A, B, and C were isolated from the mycelium culture of *Cyathus stercoreus*, and the structures of the three compounds were explained by NMR and mass spectroscopic data. Antioxidant activities of the three compounds were evaluated by the scavenging ability against ABTS⁺, DPPH, and superoxide anion radicals, and resulted that Cyathuscavins A–C showed significant antioxidant activity similar to those of reference antioxidants, BHA and Trolox. Neurodegenerative diseases, such as Alzheimer's disease and Parkinson's disease, affect millions of people worldwide [37]. Neurodegenerative diseases are happening when cells in the brain or peripheral nervous system dysfunction by time and eventually die [38, 39]. Neurotrophins promote the survival, maintenance, and regeneration of neuronal populations and can be used as a remedy against neurodegenerative diseases. Many researches in recent years,

have been focusing on natural products with the potential for treating neurodegenerative disorders [40, 41].

It is vital to search and explore novel bioactive natural products to alleviate neurodegenerative disorders. *Cyathus* species are well known as producers of bioactive cyathane diterpenoids. Ten novel cyathane diterpenoids out of 14 were isolated from the medicinal mushroom *Cyathus africanus* which was cultivated on broth medium [42]. Cyathane diterpenoids from *Cyathus* spp. showed great potentials on both neurotrophic activities and anti-neuroinflammation activities [43-46]. Yin et al., [47], reported that three novel cyathane diterpenoids stercorins A–C and two new drimane sesquiterpenoids stercorins D and E were isolated from *Cyathus stercoreus*. Stercorins A–C displayed promising neurotrophic activity in PC-12 cells and significantly suppressed LPS induced NO production in BV2 cells. Kou et al., [46], reported that, five terpenoids, including two new cyathane diterpenoids neocyathin S and neocyathin T, together with three drimane sesquiterpenoids, one known 3,6-dihydroxycinnamolide, two new ones 3,6-dihydroxycinnamolide and 2-keto-3,6-dihydroxycinnamolide, were isolated from the cultures of the basidiomycete *Cyathus africanus*. All the five compounds enhanced nerve growth factor (NGF)-mediated neurite outgrowth using rat pheochromocytoma (PC12) cells at concentration 10 μ M.

By Chemical investigation eleven new cyathane diterpenoids, designated as cyaffricanins A–K, were isolated by Yin et al., [33], from the culture broth of the basidiomycetous *Cyathus africanus* (Common name is bird's nest fungi). Their structures were explained by NMR and HRESIMS data. All compounds were investigated for their neurotrophic effect in PC-12 cells as well as their antineuroinflammatory activity in BV2 microglia cells. All of the tested diterpenoids exhibited nerve growth factor induced neurite outgrowth-promoting activity. Cyaffricanin A, exhibit anti-neuroinflammatory activity via the inhibition of pro-inflammatory enzymes expression. Also among all of them, cyaffricanin B and cyaffricanin G were the most potent neurotrophic activity. *Cyathus africanus* produced different range of cyathanes diterpenoids depending on the fermentation conditions and the period of the fermentation [48-50].

The novel polyoxygenated cyathane diterpenoids (neocyathin A–J), together with four known diterpenes, were also isolated from *Cyathus africanus* cultivated on broth medium. Those compounds were evaluated for their potential anti-neuroinflammatory activities in BV2 microglia cells. Several compounds showed differential effects on the expression of inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 in lipopolysaccharide stimulated and A 1–42 treated mouse microglia cell line BV-2. Molecular docking revealed that these bioactive compounds could interact with iNOS protein other than COX-2 protein. All isolated 14 cyathane diterpenoids together, might serve as

important lead compounds for drug discovery against neuroinflammation specially including in Alzheimer's disease [51-55].

3. CONCLUSION

Bird's nest fungi are inedible, gasteromyceteous mushrooms which are capable of secreting many new and promising secondary metabolites. These metabolites showed variable bioactivities including antimicrobial, antitumor, and anti-neuro inflammation. Information about this mushroom are still unclear. Understanding the unexplored characteristics and potentials of this tiny and fascinating mushroom is highly challenging. Moreover, further investigations, especially those describing *in vivo* studies are required in order to find out more about this interesting tiny mushroom.

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