Active Compounds from *Antrodia cinnamomea*

Subjects: Food Science & Technology | Biotechnology & Applied Microbiology

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Antrodia cinnamomea is a precious and popular edible and medicinal mushroom. It has attracted increasing attention due to its various and excellent bioactivities, such as hepatoprotection, hypoglycemic, antioxidant, antitumor, anticancer, anti-inflammatory, immunomodulation, and gut microbiota regulation properties. To elucidate its bioactivities and develop novel functional foods or medicines, numerous studies have focused on the isolation and identification of the bioactive compounds of *A. cinnamomea*.

Keywords: Antrodia cinnamomea ; bioactive ; compound ; isolation ; purification

1. Introduction

Antrodia cinnamomea (syn. *Antrodia camphorate*) is a precious edible and medicinal mushroom; it belongs to phylum Basidiomycetes, family Polyporaceae, and genus *Antrodia* [1][2]. This fungus grows only on the inner cavity of a native tree called *Cinnamomum kanehirai* Hayata at an extremely slow pace and has been known as “ruby in the forest” [3][4]. *A. cinnamomea* has been historically used to treat food and drug intoxication [5]. *A. cinnamomea* exhibits various physiological and pharmacological properties, such as anticancer, antitumor, antioxidant, anti-inflammatory, hypoglycemic, hepatoprotective, immunomodulation, and gut microbiota regulation activities [6][7][8][9]. It has also attracted increasing research attention for the development of novel medicine due to its therapeutic action, particularly its prospective application as a chemoprophylaxis agent [1].

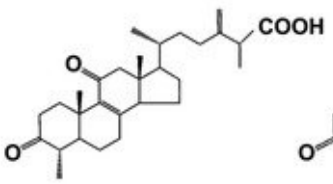
However, the wild fruiting bodies of *A. cinnamomea* are extremely expensive and in short supply because of host scarcity and slow growth. Accordingly, artificial cultivation techniques, such as cutting wood culture, solid-state fermentation, submerged fermentation, and dish culture, have been developed to supplement the expanding demand for *A. cinnamomea*. However, obtaining adequate amounts of excellent-quality *A. cinnamomea* through artificial culture has been greatly challenging [10][11]. Several differences exist between the type and content of the fruiting bodies and cultured mycelia of *A. cinnamomea*. Accordingly, numerous studies have demonstrated various methods for bridging such a disparity in bioactive metabolites between fruiting bodies and cultured mycelia [10][12].

With further in-depth studies, the research on *A. cinnamomea* is no longer limited to crude extracts. Various single substances, including polysaccharides, triterpenoids, ubiquinone derivatives, and maleic and succinic acid derivatives, have been isolated from fruiting bodies and cultured mycelia of *A. cinnamomea*. Currently, numerous kinds of isolation methods are used for the metabolites from *A. cinnamomea*. However, the standardization of product quality and purity is lacking. Numerous active substances remain undiscovered due to the limitations of isolation and purification methods.

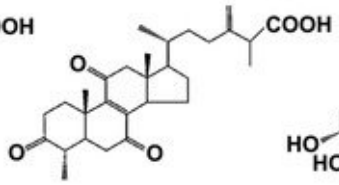
2. Isolation and Purification of Bioactive Compounds from *A. cinnamomea*

More than 200 compounds have been isolated and identified in *A. cinnamomea*, and they include polysaccharides, triterpenoids, ubiquinone derivatives, maleic and succinic acid derivatives, benzene derivatives, and glycoprotein. The chemical structures of the main bioactive components from *A. cinnamomea* are showed in **Figure 1**. However, a number of compounds with significant biological activity in *A. cinnamomea* still have not been found [13][14]. The following section summarizes the separation and purification methods of various active substances from *A. cinnamomea*.

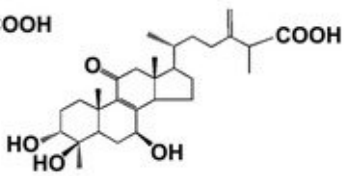
(a) Triterpenoids



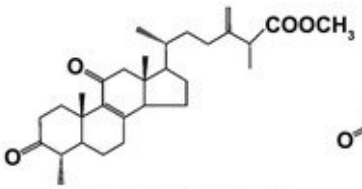
Antcin A



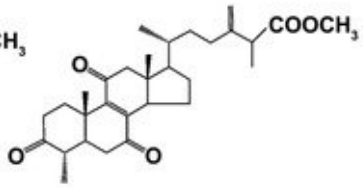
Antcin B



Antcin K

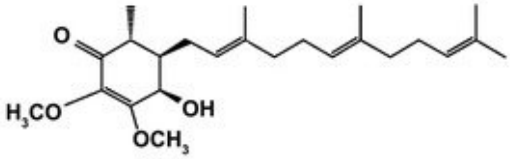


Methylantcinate A

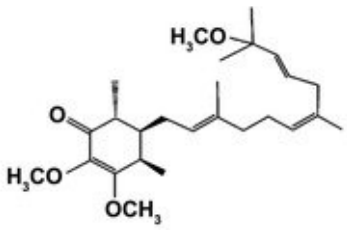


Methylantcinate B

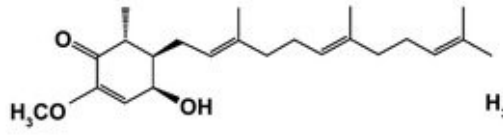
(b) Ubiquinone derivatives



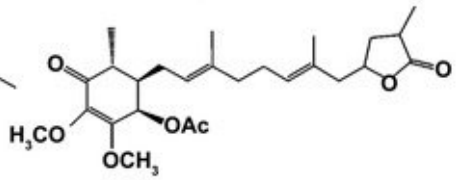
Antroquinonol



Antroquinonol C



Antroquinonol D



4−acetyantroquinonol B

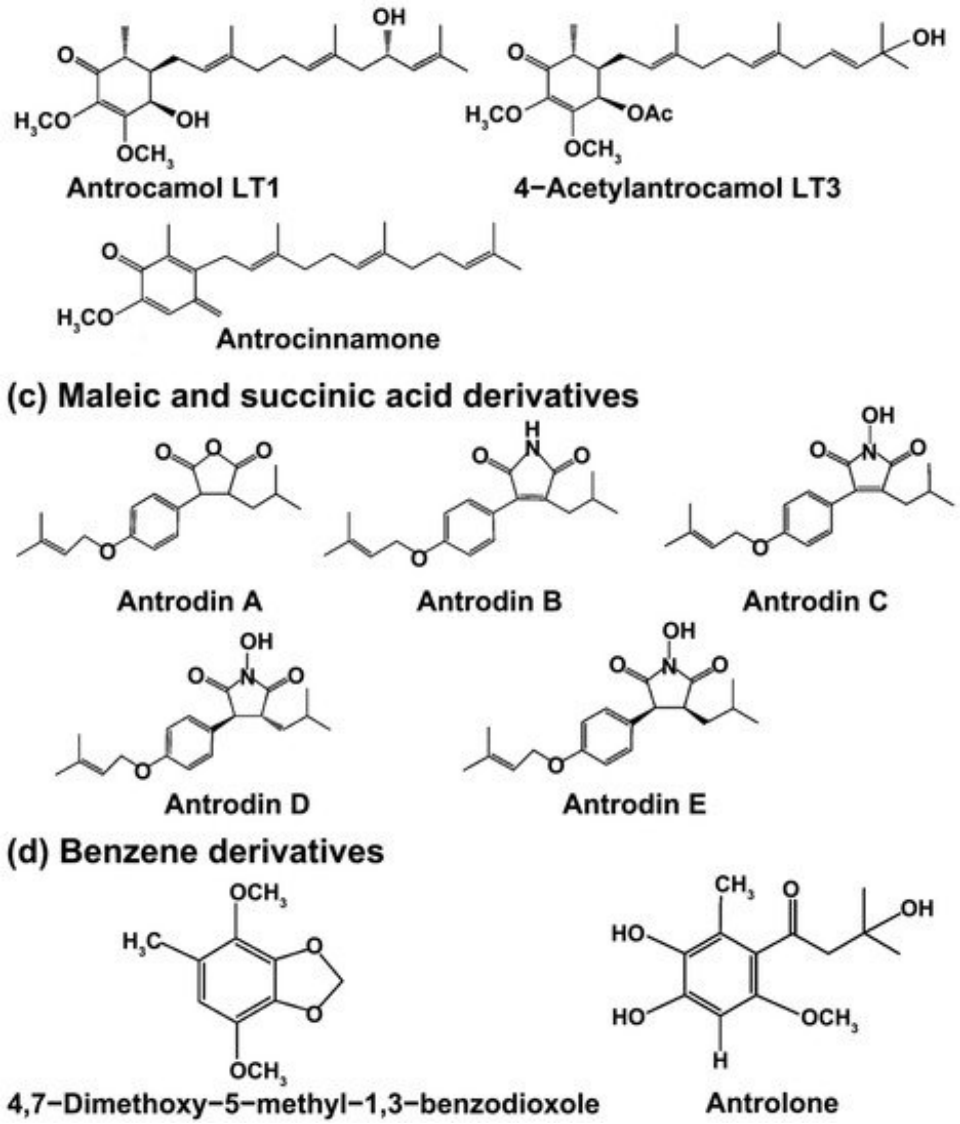


Figure 1. The chemical structures of the main bioactive components from *Antrodia cinnamomea*. (**a**) Triterpenoids, (**b**) ubiquinone derivatives, (**c**) maleic and succinic acid derivatives, (**d**) benzene derivatives.

3. Identification and Quantification of Bioactive Compounds from *A. cinnamomea*

Aside from isolation and purification, the identification of particular bioactive compounds of *A. cinnamomea* is critical. This knowledge is essential for understanding the biological activities of these metabolites and discovering new *A. cinnamomea* bioactive compounds. Typically, the characterization and quantification of natural products can use various detection techniques, including nuclear magnetic resonance (NMR), infrared spectra, and mass spectrometry (MS) detection. NMR has steadily developed and has become an essential technology for chemical identification and structural characterization. This technique enables the acquisition of data from extremely mass-limited samples and the investigation of very complex materials [15]. ^{1}H-NMR and ^{13}C-NMR analyses were used to identify compounds from solid-state cultured mycelia of *A. cinnamomea* [16]. Finally, a quinone, four phenolic acid derivatives, three ubiquinone derivatives, two alkaloids, and a triterpenoid has been identified. These compounds exhibit potent neuroprotective activities against 6-hydroxydopamine-induced toxicity in PC12 cells [16]. NMR can also be used to determine the amounts of various components in a mixture quickly and precisely. This method, known as quantitative NMR (qNMR), has found novel uses in biological and pharmacological research. qNMR is a primary ratio method compared with other instrumental analytical methods because the resulting peak areas are proportionate to the number of matching nuclei. Consequently, qNMR has been selected for the quantitative analysis of a benzenoid-rich fraction containing three primary benzenoids of *A. cinnamomea* due to its superiority to other standard chromatographic techniques in detecting the amounts of specific herbal mixture elements [17].

Fingerprint analysis is used for the thorough assessment of the quality of herbal medicines and their associated products. Recently, the HPLC fingerprinting approach supplemented by ultraviolet (UV) and photodiode array (PDA) has been widely utilized for herbal-quality testing. However, UV and PDA detectors present several problems in the isolation of target analytes from interfering impurities. Thus, these techniques involve time-consuming sample preparation and long

HPLC run times. To resolve these issues, scientists use the more sensitive liquid chromatography–tandem MS (HPLC-MS/MS), which can offer better overall information, such as molecular weight, retention time, and analyte collision fragments. Accordingly, a validated HPLC-MS/MS method for the quick and precise measurement of compounds from *A. cinnamomea* was established [18]. The method can be used to simultaneously measure seven characteristic chemicals, including antcin A, antcin B, antcin C, antcin H, antcin K, dehydroeburicoic acid, and DMB [18]. Triterpenoids are a significant bioactive component of *A. cinnamomea*, and different cultivation techniques exhibit significant variations. Qiao et al. [19] reported the contents of 10 ergostane-type and 8 landostane-type triterpenoids of *A. cinnamomea* samples derived from cutting wood culture, solid-state fermentation, submerged fermentation, and dish culture. It was determined by ultra-high performance liquid chromatography/ultraviolet (UPLC/UV) or supercritical fluid chromatography coupled with mass spectrometry (SFC/MS, for 25R/S-antcin A) within 16 min. The result showed that the 18 kinds of triterpenoids accounted for 118.2 ± 28.2, 89.4 ± 30.8, and 116.5 ± 1.1 $mg \cdot g^{-1}$ in wood-cultured fruiting bodies, wood-cultured mycelia, and dish-cultured, respectively. However, no triterpenoids were detected in the solid support cultivation or submerged fermentation samples in this research [19]. The development of products derived from *A. cinnamomea* benefits from the precise and rapid measurement of signature compounds in various samples using this method. Ultra-performance liquid chromatography quadrupole time-of-flight MS (UPLC/Q-TOF/MS) has been used to characterize and quantify mixture compounds. For the first time, the UPLC/Q-TOF/MS method was used to identify the extracts from *A. cinnamomea* and led to the discovery of 139 chemical compounds, including 102 terpenoids, 8 benzenoids, 2 purine nucleosides, and 27 other classes [20]. The development of this method has significant implications for the exploration of *A. cinnamomea* products.

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