Extraction, isolation, identification, and bioactivity of polysaccharides from *Antrodia cinnamomea*

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Abstract

Antrodia (*A*) *cinnamomea* is a precious edible and medicinal mushroom and has attracted attention because of its rare resources and unique bioactivities. The bioactive compounds from the fruit body, mycelium, and fermentation broth of *A. cinnamomea* include triterpenoids, polysaccharides, antroquinonols, benzenoids, and succinic and maleic acid derivatives. Among these, polysaccharides are very important and are high-content bioactive compounds. The *A. cinnamomea* polysaccharides (APSs) present numerous biological activities, such as antiviral, anticancer, anti-inflammatory, antivascular, immunoregulation, antioxidant, and nerve protection. However, only few studies have focused only on APSs so far. Therefore, the cultivation methods, extraction, isolation, composition, structure, biological activity, and application of APSs are summarized in this review to provide a comprehensive and convenient reference for further research and development on APSs.

Keywords: Antrodia cinnamomea; bioactivity; composition; extraction; isolation; polysaccharides

Introduction

The edible fungi can be used as food as well as medication. A number of edible fungi products have been identified as sources of healthy food supplements and drugs for numerous types of cancers in humans (Hsieh *et al.,* 2006; Lavi *et al.*, 2006; Lee *et al.*, 2005), and omics techniques have been used to investigate active ingredients obtained from edible fungi (Cao *et al.*, 2023; Geng *et al.*, 2022). A variety of bioactive components, such as terpenes, polysaccharides, maleic acids, and their derivatives, have been identified from different types of edible fungi having good therapeutics, such as anticancer, antitumor, anti-inflammatory, hypoglycemic, hypotensive, and immunoregulatory properties (Cao *et al.*, 2023; Geng *et al.*, 2022; Lavi *et al.*, 2006; Lee *et al.*, 2005). Polysaccharides are considered as the most promising pharmacologically active components (Lavi *et al.*, 2006). The polysaccharide (PS) of *Astragalus membranaceus* has excellent anticancer potential (Yang *et al.*, 2013) and can alleviate obesity, liver steatosis, neuroinflammation, and cognitive impairment (Huang *et al.*, 2017). Polysaccharides (PSs) obtained from several mushrooms, such as *Ganoderma lucidum*, *Grifola umbellata*, and *Coriolus versicolor*, scavenge free radicals (Liu *et al.*, 1997) and inhibit tumor growth (Yukawa *et al.*, 2012).

Antrodia cinnamomea (*A. cinnamomea*), with a common name of *Antrodia camphorata*, is a precious edible and medicinal mushroom that belongs to the

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phylum Basidiomycetes, family Polyporaceae, and genus *Antrodia*. It features a sporangium on the surface of the fruiting body that contains spores, an uneven surface, and a safrole odor. Based on the color of fruiting bodies, the fungi are classified as red, yellow, and white *A. cinnamomea* (Su, *et al.*, 2023). Triterpenoids and polysaccharides are the primary active components of *A. cinnamomea* (He *et al.*, 2019; Liu *et al.*, 2004). However, the wild fruiting body of *A. cinnamomea* is scarce and expensive and cannot meet market demand. Thus, the artificial cultivation of *A. cinnamomea* has become necessary. The main artificial cultivation methods of *A. cinnamomea* include basswood culture, plate culture, solid-state fermentation, and submerged fermentation (Li *et al.*, 2015; Lu *et al.*, 2014b). Submerged fermentation is the most applied cultivation method because of its short period, high production efficiency, and easy to scale-up production (Zhang *et al.*, 2019).

Dr. Huaxiang Li is engaged in the research on *A. cinnamomea* for more than 10 years, especially focusing on the submerged fermentation of *A. cinnamomea*. In 2009, it was reported for the first time that *A. cinnamomea* could produce a large number of asexual spores (i.e., arthrospores) at a late stage of submerged fermentation under appropriate environmental and nutritional conditions (Lu *et al.*, 2011). Then, in 2014, the rapid fermentation process of *A. cinnamomea* based on asexual spore inoculation was established for the first time (Lu *et al.*, 2014b). In 2015, to shorten further the production period and save production cost, an efficient repeated batch fermentation process based on asexual spore inoculation was established, which greatly improved the production efficiency of *A. cinnamomea* in submerged fermentation (Li *et al.*, 2015). In 2017, proteomics and transcriptome techniques were used to explore the molecular regulatory mechanism underlying the asexual sporulation of *A. cinnamomea* during submerged fermentation and successfully revealed the FluG-mediated asexual sporulation signaling pathway in *A. cinnamomea* (Li *et al.*, 2017). In 2022, molecular regulatory mechanisms underlying the asexual sporulation of *A. cinnamomea* induced by nutrient restriction (Li *et al.*, 2022a) and promoted by iron ion (Li *et al.*, 2023) in submerged fermentation were revealed by transcriptomics. In addition, the excellent effect of *A. cinnamomea* polysaccharides (APSs) was observed on inflammation alleviation and intestinal flora regulation in antibiotic-induced diarrheic mice (Lu *et al.*, 2022a, 2022b).

Polysaccharides are the main product of submerged fermentation of *A. cinnamomea*. Several studies reported the significantly higher content of polysaccharides and triterpenes in *A. cinnamomea* than in other similar fungi, such as *Ganoderma lucidum* and *Volvariella volvacea*, which indicates the huge development and research value of APSs (Lee *et al.*, 2014). In addition, APSs have different biological activities, such as anticancer (Wang *et al.*, 2015), anti-inflammatory (Meng *et al.*, 2012), antioxidant (Tsai *et al.*, 2007), liver protection (Liu *et al.*, 2023), and nerve protection properties (Han *et al.*, 2020). Studies on APSs usually focused on its activity *in vivo* (Liu *et al.*, 2010) or the molecular mechanism of its biological activities (Zhang *et al.*, 2018). However, no study has been dedicated to the comprehensive summary of APSs. Thus, this review summarizes the extraction, isolation, composition, structure, and biological activity of APSs and provides a comprehensive and convenient reference for the future research on APSs.

Extraction of APSs

The extraction and characterization of APSs is a difficult but an important process (Figure 1). Since polysaccharide is a polar macromolecular compound that is often soluble in water but insoluble in organic solvents, both hot water extraction (Tsai, *et al.*, 2007) and salt water extraction (Cheng *et al.*, 2009) have become quick and easy to set up common extraction techniques. Additionally, the polysaccharides can be extracted using a diluted alkali technique. However, this method has potential to destroy the structure of APSs and decrease extraction efficiency (Lu *et al.*, 2014a).

Prior to the extraction of polysaccharides, it is essential to remove the naturally occurring oil-soluble compounds by some efficient techniques, such as supercritical fluid carbon dioxide (99.5% purity) to reduce interference with polysaccharide extraction (Chen *et al.*, 2007). After removing oil-soluble impurities, crude polysaccharides are obtained by water extraction and ethanol precipitation (Li *et al.*, 2022b). Hot-water extraction can extract most of the polysaccharides from the fruiting body and mycelium of *A. cinnamomea*. Nevertheless, it is difficult to obtain polysaccharides from cell walls by water extraction. Therefore, polysaccharides can be extracted from cell walls of *A. cinnamomea* mycelia according to a modified method. Briefly, *A. cinnamomea* mycelia are successively immersed in 5%, 2%, and 1% NaCl solution (w/v) with magnetic stirring for 1 h. Then the mycelia are defatted with chloroform and methanol for 1 h (Wang *et al.*, 2021). Irrespective of the method used to extract the polysaccharide solution, it is necessary to add 70–80% concentrated ethanol, and finally centrifuge and dissolve the precipitation in hot water to obtain crude polysaccharide solution (Zhang *et al.*, 2018). However, low extraction efficiency and high solvent usage in organic solvent extraction ultimately lead to environmental degradation and pose a threat to human health. Thus, finding alternative solvents is urgently required to have a new high-quality and environment friendly extraction technology.

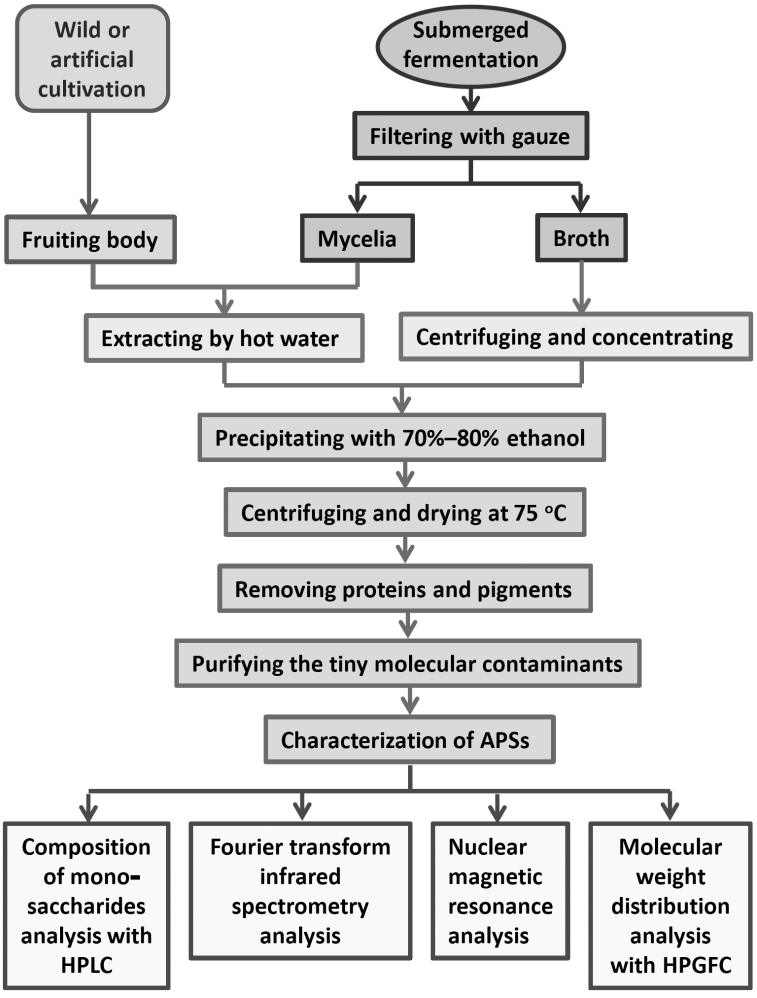

Figure 1. Flowchart of steps for the extraction, isolation, and characterization of APSs.

Crude APSs usually contain proteins, pigments, and several small molecular impurities that require further removal. Proteins are primary impurities and can be removed by the Sevag method (Zhang *et al.*, 2018), trichloroacetic acid (Liu *et al.*, 2018), and enzymatic processes (Perera *et al.*, 2017). The pigments in APSs are removed by oxidation (such as 10% H_2O_2) (Chen *et al.*, 2017), carbon decolorization (Singh *et al.*, 2013), or macroporous resin adsorption (Zou *et al.*, 2015). Subsequently, tiny molecular contaminants are typically eliminated by dialysis. Ion-exchange column chromatography and gel-column chromatography are the methods commonly used for the removal of molecular contaminants (Li *et al.*, 2022b).

In general, tiny molecular contaminants are first purified by anion-exchange column chromatography and further purified by gel column chromatography. Diethylaminoethyl (DEAE) cellulose, DEAE Sephadex, and DEAE Sepharose are the most commonly used anion exchangers, with DEAE-cellulose as the most often used (Shi, 2016). Finally, the APSs are separated by gel-column chromatography, such as Sephadex G-100, based on their size and structure (Li *et al.*, 2022b). Size-exclusion chromatography (SEC) is suitable for separating polysaccharides using aqueous solution as mobile phase and for separating water-soluble samples. It is also possible to separate and purify APS using gel filtration chromatography (GFC), which uses various salt and buffer solution concentrations as eluents. However, GFC is ineffective for mucopolysaccharide separation. Table 1 lists the reported extraction and isolation conditions for APSs.

Composition and structure of APSs

A. cinnamomea polysaccharides are classified as *A. cinnamomea* intracellular polysaccharides (AIPSs) and *A. cinnamomea* extracellular polysaccharides (AEPSs), which are derived from the mycelium/fruiting body of *A. cinnamomea* and fermentation broth, respectively. Given the complex chemical structure of polysaccharides, extracted polysaccharides are usually hydrolyzed into monosaccharides while detecting their composition

Table 1. Extraction and isolation conditions for APSs.

Sources	Extraction method	Isolation and purification method	Research results	References
Mycelia	Extracted with 80°C water at a 1:100 (w/w) ratio for 6 h	Separated by SEC	All the polysaccharides from six medicinal fungi showed no toxicity to endothelial cells up to a concentration of 250 μg/mL. The high-molecular weight (MV) polysaccharides in the size range of 2693–2876 kDa and middle-molecular weight polysaccharides in the size range of 304–325 kDa presented good antiangiogenic effects	Chen *et al.* (2005)
Mycelia	Reflux extracted thrice with 2-L double-distilled water at 90°C with constant stirring at 400 rpm for 2 h	Separated by decantation, and 3 mL of sample (APSs extract) was eluted with 0.05-N NaOH (containing 0.02% NaN_3) solution on a Sephadex G-100 column (2.5 × 100 cm^2) at a flow rate of 0.5 mL/min	The distribution of mean molecular mass of fractionated polysaccharides was in the range of 394–940 kDa. The proximate compositions from APSs fraction revealed that all fractions belonged to the category of glycoprotein, having carbohydrate–protein proportion ranging from 0.29 to 10.79 (w/w)	Chen *et al.* (2007)
Mycelia	Extracted with deionized water (2 L) at 30°C for 24 h or at 95°C for 6 h	Purified by a DEAE-cellulose ion-exchange column	PEF-1, PEMC-1, and PEMH-1 are the major water-soluble polysaccharides in the APSs named ACSC. PEMC-1 and PEMH-1 are protein-containing glycan, while PEF-1 is free of any peptide chain	Tsai *et al.* (2007)
Fruiting body	Extracted with distilled water (100 mL) at 100°C for 1 h	Separated into six fractions by Amicon Ultra-15 5-, 10-, 30-, 50-, and 100-K centrifugal filter devices (Millipore, County Cork, Ireland)	Four different molecular weights (<5, 5–30, 30–100, and >100 kDa) of APSs were obtained. The APSs of MV >100 kDa substantially and dose-dependently reduce the development of neovascularization *in vitro*	Yang *et al.* (2009)
Fruiting body	Extracted with boiling water at a ratio of 1:25 (w/v) for approximately 8–12 h	Passed through a Sephadex G50 (Amersham Pharmacia Biotech, Piscataway, NJ, USA) gel filtration column and further purified by an anion-exchange column of DEAE-cellulose	Percentage of APSs in the lyophilized extracts of fruiting bodies was more than 98%. The APSs can reduce the expressions of inflammatory mediators at the injured site and circulation, especially in the late stage of sepsis	Meng *et al.* (2012)
Mycelia	Extracted with cold water	Separated by SEC (90 cm H × 1.6 cm D) at a flow rate of 0.4 mL/min	The obtained APSs significantly enhance the phagocytosis and bactericidal activity of J774A.1 murine macrophages against *Escherichia coli*	Perera *et al.* (2017)
Mycelia	Extracted with boiled water for 3 h	Purified by DEAE-52 cellulose (2.6 × 20 cm^2) and Sephadex G-100 column chromatography (1.1 × 100 cm^2)	The APSs named ACPS-1 with a MV of 22.96 kDa was obtained and presented an outstanding anticancer activity against cervical and skin cancer cells	Zhang *et al.* (2018)
Mycelia	0.1-M sodium acetate (pH 5.5) containing 5-mM cysteine, 100-mg papain, and 5-mM ethylenediaminetetraacetic acid (EDTA) at 60°C for 24 h	Purified by GFC	The obtained APSs show anticancer effects by inhibiting the EGFR/ERK signaling pathway	Lu *et al.* (2021)
The cell wall of mycelia	5%, 2%, and 1% NaCl solution (w/v) with magnetic stirring for 1 h, defatted with chloroform and methanol for 1 h, and extracted with distilled water in a boiling water bath for 3 h	Purified by cross-flow ultra-filtration with molecular weight (MW) cut-off of 100 kDa (Pelli-con®XL, Millipore, USA)	Only 3.82% of mushroom cell wall (MCW) drymatter could be extracted by hot water. About half (49.18%) of the MCW was composed of alkali soluble fractions, while mycelia cold alkali-extracted fraction harvested (39.13%) significantly higher than mycelia hot alkali-extracted fraction	Wang *et al.* (2021)

SEC: size-exclusion chromatography; DEAE: diethylaminoethyl; EGFR: epidermal growth factor receptor; ERK: extracellular signal-regulated kinase; GFC: gel filtration chromatography.

by high-performance liquid chromatography (HPLC) (Mendes *et al.*, 2020), thin-layer chromatography (TLC) (Kwamla and Thomas, 2022), gas chromatography (GC) (Andres *et al.*, 2020), and high-performance anion-exchange chromatography (HPAEC-AED) (Brunt *et al.*, 2020). TLC has low sensitivity and poor separation effect; HPAEC-AED is expensive, and the incomplete degradation of polysaccharides by acid hydrolysis will affect the quantitative analysis. Thus, HPLC and GC are widely used due to their fast separation speed, good separation effect, and good reproducibility.

Su *et al*. (2016) analyzed the polysaccharides from the fruiting body of *A. cinnamomea* (collection number: KJ-AC-14) by the 1-phenyl-3-methyl-5-pyrazolone-precolumn derivatization method and observed that the monosaccharide composition of APSs includes fucose (2.0 ± 0.1 molar%), galactose (2.9 ± 0.2 molar%), glucose (84.0 ± 1.7 molar%), mannose (7.2 ± 1.9 molar%), rhamnose (1.4 ± 0.3 molar%), and xylose (2.4 ± 0.2 molar%). Cheng *et al*. (2018) analyzed the AIPSs from strain B86 (Taipei Institute of Forestry, Taiwan) in solid-state fermentation by HPAEC and observed that it consists of fucose (3.08 ± 0.05 μmol/g), glucosamine (8.58 ± 0.10 μmol/g), galactose (35.89 ± 0.67 μmol/g), glucose (600.42 ± 7.20 μmol/g), and mannose (20.24 ± 0.73 μmol/g). Zhang *et al*. (2018) isolated a polysaccharide, named ACPS-1, from the submerged fermentation broth of *A. cinnamomea*. Then the authors analyzed the structure of ACPS-1 by DEAE-52 Sephadex G-100 column chromatography, Fourier transform infrared spectroscopy (FTIR), and nuclear magnetic resonance (NMR), and discovered that it consisted of mannose, glucose, fucose, xypyranose, arabinose, fructose, and rhamnose at a molar ratio of 31.27:1.77:1.44:1.34:1.00, and its backbone consisted of repeating α and →3), its→6), its →2), and backbone glycosidic linkages.

The composition of AIPSs and AEPSs from the ATCC 200183 strain in submerged fermentation at 26°C and 150 r/min for 12 days was analyzed by the phenol–sulfuric acid method (Lu *et al.*, 2022a, 2022b). Finally, it was observed that the AIPS consisted of galactose (28.52%), glucose (55.31%), mannose (14.34%), and galactosamine (1.83%) whereas the AEPS comprised glucose (84.73%), galactose (7.84%), mannose (5.27%), galacturonic acid (0.76%), and glucuronic acid (1.4%). Table 2 lists the reported monosaccharide compositions of APSs.

The structure of APSs was detected using FTIR, hydrogen NMR (H-NMR), carbon NMR (C-NMR), and infrared radiation. Su *et al*. (2016) analyzed the structure of APSs from the fruiting body of *A. cinnamomea* by FTIR and observed the presence of pyranose. Zhang *et al*. (2018) analyzed the structure of APSs from the mycelium of *A. cinnamomea* in submerged fermentation by FTIR and revealed the presence of pyranose. Lu *et al*. (2021) explored the structure of a APS type (Na10_SPS-F3) using H-NMR and C-NMR, and reported that it is a 3-o-sulfated malonosaccharide, whose pentose 1,4-β-glc is linked to hexose 1,4-α-glc.

Liu *et al*. (2017) analyzed the structure of an APS type, named ACW0, by one-dimensional (1D) and 2D NMR and observed that it was a type of mannose–galactan whose main chain was α-d-1,6-Gal. Almost one nonreducing terminal α-D-Man and α-L-Fuc in every six α-d-1,6-Gal residues was attached to C-2. In addition, we analyzed the structures of AIPS and AEPS from the ATCC 200183 strain in submerged fermentation by FTIR

Table 2. Monosaccharide compositions of APSs.

Strains	Sources	Cultivation mode	Monosaccharide composition	References
Collection number: KJ-AC-14	Fruiting body	-	Fucose (2.0 ± 0.1%), galactose (2.9 ± 0.2%), glucose (84.0 ± 1.7%), mannose (7.2 ± 1.9%), rhamnose (1.4 ± 0.3%), and xylose (2.4 ± 0.2%)	Su *et al.* (2016)
Taipei Institute of Forestry: B86	Mycelium	0.5-mM potassium sulfate, 24-g/L potato-dextrose-broth (PDB), and 20-g/L glucose for 49 days	Fucose (3.08 ± 0.05 μmol/g), glucosamine (8.58 ± 0.10 μmol/g), galactose (35.89 ± 0.67 μmol/g), glucose (600.42 ± 7.20 μmol/g), and mannose (20.24 ± 0.73 μmol/g)	Cheng *et al.* (2018)
-	Mycelium	Submerged culture for 10 days	Mannose, glucose, fucose, xypyranose, arabinose, fructose, and rhamnose at a molar ratio of 31.27:1.77:1.44:1.34:1.00	Zhang *et al.* (2018)
ATCC 200183	Mycelium	Immersed for 12 days at 150 r/min at 26°C	Galactose (28.52%), glucose (55.31%), mannose (14.34%), and galactosamine (1.83%)	Lu *et al.* (2022b)
ATCC 200183	Broth	Screened with four layers of gauze after immersion for 12 days at 150 r/min at 26°C	Glucose (84.73%), galactose (7.84%), mannose (5.27%), galacturonic acid (0.76%), and glucuronic acid (1.4%)	Lu *et al.* (2022a)

Note: "-" not mentioned.

Table 3. Structural analysis of APSs.

Strains	Sources	Method	Structure	References
Collection number: KJ-AC-14	Fruiting body	FTIR	Pyranose ring	Su *et al.* (2016)
-	Mycelium	FTIR and NMR	The main chain of ACW0 polysaccharide is 1,6-Gal, and the branch is located at the C-2 site of 1,2,6-Gal	Liu *et al.* (2017)
-	Mycelium	FTIR	Pyranose ring	Zhang *et al.* (2018)
Taipei Institute of Forestry: B86	Mycelium	NMR	3-O-sulfomalonyl glucan with eight 1,4-β-Glc moieties connected with ten 1,4-α-Glc moieties	Lu *et al.* (2021)
ATCC 200183	Broth	FTIR	β-type glucoside	Lu *et al.* (2022a)

Note: "-" not mentioned.

and revealed that AEPS was a beta-type glucoside with a pyranose ring whereas AIPS possessed (-C≡C-H) and (C-O) functional groups (Lu *et al.*, 2022a, 2022b). Table 3 lists the results of the structural analysis of APSs. Figure 1 presents the steps involved in the extraction, isolation, and characterization of APSs.

Bioactivity of APSs

The content of APSs in *A. cinnamomea* is significantly higher than that of similar fungi, such as *Ganoderma lucidum* and *Volvariella volvacea*; this indicates that APSs may possess different bioactivities with great research and development values (Hseu *et al.*, 2002; Song and Yen, 2002).

Safety of APS

Chen *et al.* (2005) evaluated the toxicity of APS toward endothelial cell (EC) viability and found that APS showed no toxicity to endothelial cells up to a concentration of 250 μg/mL. Tsai *et al.* (2007) found that all cellular viabilities were higher than 90% if Chang liver cells were treated with APS at concentration of up to 200 μg/mL at 37°C for 24 h, indicating that APS was not cytotoxic to Chang liver cells. Wang *et al.* (2015) found that serum TGF-β quantity in common mouse was 39.59 ± 5.645 ng/mL, compared to the mice fed with *A. cinnamomea* *β*-glucan at 32.8 ± 1.879 ng/mL. There was no significant difference between the two groups. It was obvious that daily oral intake of *A. cinnamomea* *β*-glucan does not alter serum TGF-β in normal mice. Zhang *et al.* (2018) assessed the safety of APSs by monitoring the proliferation of normal mouse spleen cells treated with APSs at concentrations ranging from 25 g/mL to 1000 g/mL; the authors observed that APSs showed no effect on cell viability at any concentration. Moreover, in several cases, the APSs increased cell viability. Thus, APSs were safe enough and noncytotoxic toward normal cells.

Anticancer activity of APSs

To date, the impact of different cancers, such as lung, liver, and breast cancers, on the health of patients is increasingly aggravating. With increase in the incidences of different cancers, the drugs used to treat cancers are also improving constantly. Fungal polysaccharides have been identified as pharmacologically active antitumor components (Wang *et al.*, 2015). *A. cinnamomea*, as a valuable edible fungus, has active compounds with excellent anticancer effects. *In vitro* and *in vivo* studies have revealed that the polysaccharides from the fruiting bodies and mycelium of *A. cinnamomea* have potent anticancer properties (Ho *et al.*, 2008; Liu *et al.*, 2004).

DNA damage is a characteristic of cancer cells, but it can also be a target for treatments that fight the disease. Zhang *et al.* (2018) mentioned that a new polysaccharide (ACPS-1) isolated from *A. cinnamomea* mycelium could cause apoptosis and cell cycle arrest in cervical and skin cancer cells by obstructing the DNA repair pathway controlled by topoisomerase I/tyrosine DNA phosphodiesterase I. Fa *et al.* (2015) used Lewis lung cancer cells, a highly aggressive mouse lung cancer cell line, to investigate the antimetastatic activity of antrodan (a glycoprotein isolated from the mycelium of *A. cinnamomea*) by direct or indirect immunomodulatory effects. The results showed that direct and indirect immunomodulatory effects had anti-metastasis potential in mouse lung cancer cells. In addition, at the same concentration (50 and 60 μg/mL), antrodan exhibited a stronger indirect immunomodulatory effect on tumor metastasis than direct effect.

Replacement of several functional groups of polysaccharides with sulfate groups could improve their biological activity (Wang *et al.*, 2013, 2018). Lu *et al.* (2017a) observed that sulfated polysaccharides (SPS) from *A. cinnamomea* not only inhibited the survival ability of lung cancer cells but also reduced the expression of transforming growth factor β receptor (TGFR) protein and blocked the intracellular signaling pathway regulated

by TGFR, which lead to the migration of lung cancer cells. Moreover, the authors were the first to isolate and identify a 1,4-D-galactoman (B86-III) containing 1,6-branched chains from *A. cinnamomea*. TGFR and its downstream signals, focal adhesion kinase (FAK), and Slug were involved in the development of lung tumors (Lu *et al.*, 2017b). B86-III inhibited the expression and migration of Slug by downregulating the TGFR I protein and inhibiting the phosphorylation of FAK. Finally, the activity of H1975 lung cancer cells was inhibited (Lu *et al.*, 2017b).

Lu *et al.* (2018) identified an anticancer sulfated β-(1→4)-D-glucan (denoted as AC-SPS-F3) with long β-(1→6)-Glcp branches and a very high sulfate ratio from *A. cinnamomea*. It inhibited the action of epidermal growth factor receptor (EGFR) and mammalian target of rapamycin. Furthermore, using gel column chromatography, Lu *et al.* (2021) further isolated SPS and obtained Na10_SPS-F2 and Na10_SPS-F3, which reduced cell viability by eliciting apoptotic responses. Specifically, Na10_SPS-F3 showed anticancer effects by inhibiting the EGFR/extracellular signal-regulated kinase (ERK) signaling pathway. Lu *et al.* (2023) discovered a sulfated galactoglucan (3-SS) in *A. cinnamomea.* It could impair the proliferation of H1975 lung cancer cells through EGFR/ERK/Slug signaling.

In addition, the polysaccharides isolated from *A. cinnamomea* mycelium (AC-PS) significantly inhibited the proliferation of leukemia U937 cells by activating mononuclear cells (MNCs) (Liu *et al.*, 2004). In their recent study, Lin *et al.* (2023) discovered that SPS from *A. cinnamomea* with a molecular weight (MW) of 7.9 kDa (dubbed *ZnF3* gene) had dual anticancer properties, killing cancer cells while stimulating macrophages. *ZnF3* downregulated the expression of transforming growth factor beta receptor (TGFβ) in lung cancer cells. In parallel, *ZnF3* activated macrophages via induction of tumor necrosis factor-α (TNF-α) and interleukin 6 (IL-6) secretion, nitric oxide (NO) production, and phagocytosis. *ZnF3* activated the serine/threonine kinase–rapamycin (AKT/mTOR) pathway and induced M1 type macrophage polarization. Cancer cells co-cultured with *ZnF3*-stimulated macrophages lead to the inhibition of lung cancer cells.

The main disadvantages of natural compounds are their low solubility and bioavailability, and inability to adhere to prescribed treatments. Nanotechnology is one of the new tools used for diagnosis, treatment, and prevention of cancer (Siddiqui *et al.*, 2012). Therefore, nanotechnology offers another mode to improve the bioavailability of naturally active food ingredients. Kong *et al.* (2013) successfully prepared silica or silica chitosan nanoparticles coated with APSs. The 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide experiments showed that APSs coated with silica or silica chitosan nanoparticles inhibited the growth of 10 different human cancer cell lines but were nontoxic to three normal human cell lines. The APSs encapsulated in silica or silica–chitosan nanoparticles showed better stability and increased antitumor activity. Table 4 lists the reported anticancer activity of APSs.

Hepatoprotective activity of APSs

The active compounds from *A. cinnamomea* are used for the treatment of liver injury, alcoholic liver, nonalcoholic fatty liver disease, and liver cancer with excellent therapeutic effects (Ruan *et al.*, 2022). APSs can considerably protect the liver. Lee *et al.* (2002) revealed the good anti-hepatitis B virus (HBV) effects of AC-PS. Ho *et al.* (2008) observed that the AC-PS had a good protective effect against ethanol-induced liver injury. Tang *et al.* (2019) isolated a galactosylglucan named ACP2 from *A. cinnamomea* mycelium and evaluated its anti-inflammatory effects on human L02 (liver cell line) cells. The results showed that ACP2 drastically alleviated

Table 4. Anticancer activity of APSs.

Sources	Cancer type	Optimum concentration	References
Mycelium	Leukemic	100 μg/mL	Liu *et al.* (2004)
Mycelium	Lung cancer	50 μg/mL for migration inhibition and 60 μg/mL for invasion inhibition	Fa *et al.* (2015)
Mycelium	Lung cancer	200 μg/mL	Lu *et al.* (2017a)
Mycelium	Lung cancer	200 μg/mL	Lu *et al.* (2017b)
Mycelium	Cervical cancer	1,000 μg/mL	Zhang *et al.* (2018)
	Skin cancer	800 μg/mL	
Mycelium	Lung cancer	550 μg/mL	Lu *et al.* (2018)
Mycelium	Lung cancer	800 μg/mL	Lu *et al.* (2021)
Mycelium	Lung cancer	>400 μg/mL	Lin *et al.* (2023)
Mycelium	Lung cancer	-	Lu *et al.* (2023)

endotoxin-induced hepatocyte inflammation by decreasing the expressions of cyclooxygenase-2, IL-1b, TNF-α, and IL-6. Han *et al.* (2006) isolated a neutral polysaccharide, named ACN2a, from *A. cinnamomea* mycelium and used it (gavaged 0.4 and 0.8 g/kg/day) to treat liver injury in mice induced by *Propionibacterium acnes* and lipopolysaccharides (LPS). The results showed that ACN2a could considerably suppress rise in serum aspartate aminotransferase and alanine aminotransferase activities produced by *P. acnes* and LPS in mice and presented a liver protective effect.

In addition, overexpression of reactive oxygen species (ROS) in liver damage leads to inflammation (Matsuzawa *et al.*, 2005). Yang *et al.* (2022) reported that APSs could activate the NF-E2-related factor 2 signaling pathway (an important antioxidant signaling pathway). APSs could also inhibit ROS expression, increase the gene expression of superoxide dismutase (SOD), and ultimately reduce the expression of inflammatory cytokines. In general, APSs effectively ameliorated liver damage in mice. Moreover, abnormal expression of nucleotide-binding oligomerization domain (NLRP3; leucine-rich repeat and pyrin domain-containing protein 3, a kind of inflammasome) has been found in various models of liver injury (Duffield *et al.*, 2005). Ruan *et al.* (2022) proved that APSs could activate the autophagy of Kupffer cells and induce the degradation of NLRP3, which reduced inflammatory response, inhibited the release of inflammatory factors, and provided liver protection. Table 5 lists the reported hepatoprotective activity of APSs.

Anti-inflammatory activity of APSs

Meng *et al.* (2012) used APSs to treat the sepsis caused by cecal ligation and puncture in mice, and observed that both polysaccharides from the mycelium and fruiting bodies of *A. cinnamomea* could reduce the expression of inflammatory mediators at the injured site and circulation, especially in the late stage of sepsis. However, the polysaccharides from the fruiting bodies of *A. cinnamomea* were more effective than those from the mycelia of *A. cinnamomea* in lowering inflammatory response. In addition, Wu *et al.* (2007) demonstrated that an alkaline extraction-isoelectric precipitation fraction in APSs (AC-2) could decrease the synthesis of IL-6, IL-10, monocyte chemoattractant protein (MCP)-5 and NO in LPS-stimulated mouse macrophages. This transcriptional downregulation of *IL-6*, *IL-10*, and *inducible NO synthase* (*iNOS*) genes resulted in the inhibition of IL-6, IL-10, and iNOS. Zheng *et al.* (2017) obtained an ACHO (AC polysaccharides at 90°C, an oligosaccharide product) from ACP at 90°C by trifluoroacetic acid degradation and observed that ACHO dramatically reduced the inflammatory reactions induced by LPS *in vitro* and *in vivo* by suppressing the expression levels of mRNA for several pro-inflammatory cytokines, including IL-6, IL-8, IL-1, TNF-α, and monocyte chemoattractant protein (MCP-1). The underlying molecular mechanism of ACHO's anti-inflammatory effect promoted O-GlcNAcylation, which in turn prevented the phosphorylation of p38 mitogen-activated protein kinase and protein kinase B (AKT).

Cheng *et al.* (2016) isolated polysaccharides and SPS from *A. cinnamomea* mycelium and used the RAW264.7 macrophages induced by LPS to study the effect of polysaccharides and SPS on inflammatory response. The authors observed that SPS significantly inhibited the release of TNF-α and IL-6 more than non-sulfate polysaccharides, with 100% inhibition. This result indicated that the degree of sulfation of APSs could have a role in anti-inflammatory action. Chiu *et al.* (2013) further purified APSs by Sepharose CL-6B column chromatography and obtained antrodan. Then the authors showed that antrodan was completely harmless to the RAW 264.7 cell line at doses as high as 400 μg/mL and could reduce the inflammatory response induced by LPS in RAW 264.7 cell line. In addition, antrodan significantly reduced NO production at a low dose of 18.75 μg/mL. This finding indicated that several oligosaccharides with less polymerization typically exhibited better solubility in water, compared to polysaccharides (McCranie *et al.*, 2014). Peng *et al.* (2015) observed that a unique β-glucan (antrodan) from *A. cinnamomea* mycelium could be used to treat benign prostatic hyperplasia (BPH). Moreover, kidney and testicular apparent weight was completely unaffected by antrodan, which implied that antrodan therapy was safe.

Table 5. Hepatoprotective activity of APSs.

Sources	Hepatoprotective type	Optimum concentration	References
Mycelium	HBV	1,000 U/mL	Lee *et al.* (2002)
Mycelium	Propionibacterium acnes and LPS-induced hepatic injury	0.8 g/kg/day	Han *et al.* (2006)
Mycelium	Alcohol-induced liver injury	500 mg/L	Ho *et al.* (2008)
Mycelium	LPS-induced hepatocyte inflammation	100 μg/mL	Tang *et al.* (2019)
Mycelium	Inflammatory response in liver injury	15 mg/L	Yang *et al.* (2022)
Mycelium	Liver injury caused by abnormal NLRP3 expression	15 mg/kg	Ruan *et al.* (2022)

The effects of AIPS and AEPS on inflammation and intestinal flora disturbance induced by lincomycin hydrochloride (LIH) in mice were investigated in the past studies (Lu *et al.*, 2022a, 2022b). Finally, it was observed that AIPS considerably decreased weight loss, restored immunological organ indices, and markedly reduced the levels of pro-inflammatory cytokines TNF-α and IL-6 in mouse serum. Similarly, AEPS could significantly lessen immune organ damage and lower serum levels of inflammatory factors, such as IL-6 and TNF-α. In addition, AIPS and AEPS could regulate and restore the gut microflora structure in mice by significantly reducing the relative abundance of intestinal harmful microorganisms and improving the relative abundance of intestinal beneficial microorganisms.

Lu *et al*. (2023) discovered a sulfated galactoglucan (3-SS) in *A. cinnamomea*. The anti-inflammation effects of 3-SS on RAW264.7 macrophage cells, such as IL-6 inhibition, restoration of LPS-induced IκB protein degradation, and inhibiting LPS-induced TGFRII protein degradation, were confirmed to occur via AKT, ERK1/2, and p-38. Table 6 lists the reported anti-inflammatory activity of APSs.

Neuroregulatory effects of APSs

The degeneration and necrosis of dopaminergic neurons have been linked to neuroinflammation, which has been demonstrated to persist in Parkinson's disease (Hirsch *et al.*, 2012). The polysaccharides from several medicinal and edible fungi or plants, such as *Astragalus membranaceus* and *Lentinula edodes*, exert good neuroprotective benefits or inhibitory effects on neuroinflammation and present great potential for clinical application (Huang *et al.*, 2017; Iancu *et al.*, 2005). APSs also present good neuroprotective properties and are effectively used in the treatment of Alzheimer's disease (Wang *et al.*, 2012). Although *A. cinnamomea* mycelium contained more polysaccharides than the fruiting body, the polysaccharides from its fruiting body are more potent and have a higher inhibitory effect on cytotoxicity (Wang *et al.*, 2012).

As a neurotoxin, 6-hydroxydopamine (6-OHDA) can be selectively absorbed by dopaminergic neurons of monoamine oxidase and transformed into free radicals that harm nerve cells and cause lesions similar to those in Parkinson's disease (Zhao *et al.*, 2016). Han *et al*. (2020) reported that APSs enhanced 6-OHDA expression in a rat model of Parkinson's disease induced by 6-OHDA. However, APSs did not affect the level of serotonin transmitter but mainly improved the survival and regeneration of dopaminergic neurons. APSs also lowered the NLRP3 expression in inflammasome and completely inhibited the expression of NLRP3 inflammatory body and its inflammatory components. In addition, 6-OHDA may activate ROS-NLRP3 to cause death of dopaminergic neurons; however, APSs can suppress this signal and save dopaminergic neurons (Han *et al.*, 2020). Ultimately, APSs improved the neurobehavior, such as mobility and coordination, of mice with Parkinson's disease (Han *et al.*, 2019).

Antivascular activity of APSs

Angiogenesis, the growth of new blood vessels from those that already exist, largely occurs during human development and reproduction. Vascular abnormalities are caused by the imbalance between the regulation of angiogenesis boosting and inhibiting factors (Cuvillier, 2017). Endothelial cell migration, proliferation, and tube formation are important processes in angiogenesis. Cancer cells produce large amounts of vascular endothelial growth factor (VEGF), basic fibroblast-like growth factor, IL-8, and transforming growth factor-H, all of which promote recruitment and proliferation of endothelial cells (Ferrara, 2000).

Table 6. Anti-inflammatory activity of APSs.

Sources	Inflammatory type	Optimum concentration	References
Mycelium	LPS-induced gene activation in mouse macrophages	200 mg/L	Wu *et al.* (2007)
Fruiting body and mycelium	Polymicrobial sepsis	100 mg/kg	Meng *et al.* (2012)
Mycelium	LPS-induced inflammation	18.75 μg/mL	Chiu *et al.* (2013)
Mycelium	Benign prostate hyperplasia	30 mg/kg	Peng *et al.* (2015)
Mycelium	LPS-induced inflammation	500 μg/mL	Cheng *et al.* (2016)
Mycelium	LPS-induced inflammation	100 μg/mL	Zheng *et al.* (2017)
Broth	LIH-induced inflammation	0.25 g/kg	Lu *et al.* (2022a)
Mycelium	LIH-induced inflammation	0.25 g/kg	Lu *et al.* (2022b)
Mycelium	Macrophage cells-induced inflammation	-	Lu *et al.* (2023)

LIH: lincomycin hydrochloride; LPS: lipopolysaccharides.

In order to study the link between the structure and functioning of polysaccharides on angiogenesis, Chen *et al*. (2005) analyzed polysaccharides from six medicinal fungi, including *A. cinnamomea*, *Antrodia malicola*, *Antrodia xantha*, *Antrodiella liebmannii*, *Agaricus murrill*, and *Rigidoporus ulmarius*, and observed an association between effect on angiogenesis and molecular size of polysaccharides. In brief, the medium- and high-molecular weight polysaccharides with a respective size range of 304–325 and 2693–2876 kDa exhibited good antiangiogenic effects. In addition, APSs have a very high content of β-D-glucan (Su *et al.*, 2016), which has (1,3)-β-glucans with (1,6)-β-linked side chains (Lee *et al.*, 2002). The β-glucan from *A. cinnamomea* shows no genotoxicity (Chen *et al.*, 2018). Cheng *et al.* (2011) isolated a subcomponent of APS named B85PS-I-V, whose main component is 1,6-α-D-mannogalactan (B85PS-III-1) that consists of galactose and caramel. The authors observed that B85PS-III-1 could inhibit the angiogenesis-related processes of endothelial cell migration and tube formation. Cheng *et al*. (2005) also observed that APSs reduced the production of cyclin D1 by blocking the VEGF receptor signaling pathway, hence reducing angiogenesis.

Yang *et al*. (2009) also investigated the immunomodulatory effects of APSs on angiogenesis and revealed that the APSs with MW > 100 kDa substantially and dose-dependently reduced the development of neovascularization *in vitro*. Moreover, the authors revealed that APSs inhibited the angiogenesis actually via decreasing VEGF secretion in tumor cells and activating mononuclear cells (MNCs) to release abundant IL-12 and interferon-γ (IFN-γ). In addition, SPS presented excellent anti-cancer and anti-inflammatory effects (Lin *et al.*, 2019, 2020). Cheng *et al*. (2018) stated that SPS considerably inhibited the formation of vascular endothelial cell tubes. Liu *et al*. (2017) isolated a heterogalactan, named ACW0, from *A. cinnamomea* and discovered that its sulfated derivative, ACW0-Sul, with sulfate substitutions at C-3 and C-4 of 1,2,6-linked galactose, markedly inhibited the tube formation and motility of human microvascular endothelial cells (HMEC-1). Table 7 lists the reported antivascular activity of APSs.

Immune regulation activity of APSs

A. cinnamomea polysaccharides play a good role in immune regulation, especially in promoting T cell activation (Liu *et al.*, 2010). The immune system is a crucial defense mechanism that can identify and eliminate foreign bodies, such as viruses and harmful microbes, identify cancerous cells, and keep the body stable (Miao *et al.*, 2015). The immunological aging process is epidemiologically linked to the majority of common aging-related problems (Gress and Deeks, 2009).

Sheu *et al*. (2009) isolated a polysaccharide, named ACA, from *A. cinnamomea* mycelium and observed that ACA was a glycoprotein that could activate macrophages through a toll-like receptors/myeloid differentiation primary response protein (TLR2/MyD88)-dependent mechanism, which directly improved macrophage activity. Chen *et al*. (2008) investigated the effect of APSs on immune functioning by directing cytokine expression and splenic cell immunological modulation in *Schistosoma mansoni*-infected mice. After 2, 4, and 6 weeks of oral treatment, the mice exhibited high mRNA expression levels of IFN-γ and TNF-α *in vivo* and increased levels of immunological factors in spleen cells. In addition, the APSs prevented *S. mannii* infection in BALB/c mouse model. Liu *et al*. (2018) revealed that APSs enhanced cyclophosphamide (CTX)-induced immunosuppression in BALB/c mice. In brief, 4 weeks of oral APS treatment improved body weight, organ index, T cell performance, and natural killer cell cytotoxicity of mice. The APSs also successfully boosted the overall antioxidant capacity by promoting the activities of SOD, catalase, and glutathione peroxidase in the serum and spleen and by preventing the rise of ROS and malondialdehyde levels.

Asthma is a chronic disease characterized by airway inflammation caused by dysregulation of cytokines secreted by allergen-specific type 2 T-helper cells. Dendritic cells (DCs) act as both initiator of immune response and inducer of T cell tolerance (Banchereau and Steinman, 1998; Pulendran *et al.*, 2001). Liu *et al*. (2010) studied the immunomodulatory effects of polysaccharides, named GF2, from *A. cinnamomea* on dendritic cells and its capability to prevent ovalbumin-induced asthma

Table 7. Antivascular activity of APSs.

Sources	Antivascular type	Optimum concentration	References
Mycelium	VEGF receptor phosphorylation and interaction with VEGF	0.46 μg/mL	Cheng *et al.* (2005)
Mycelium	Inhibition of angiogenesis	100 μg/mL	Yang *et al.* (2009)
Mycelium	Inhibition of angiogenesis	7.07 μg/mL	Cheng *et al.* (2011)
Mycelium	Inhibition of tube formation and motility of HMEC-1 cells dose dependently	3.5 μM	Liu *et al.* (2017)
Mycelium	Inhibition of the formation of vascular endothelial cell tubes	160.92 μg/mL	Cheng *et al.* (2018)

in an allergic asthma mouse model, and observed that GF2 could be utilized as an adjuvant to prevent the development of allergic asthma by developing immunological tolerance. Lin *et al.* (2015) further studied the effect of high-molecular weight APS (hmwAPS) on immune functioning of dendritic cells and revealed that hmwAPS promoted the production of proinflammatory cytokines and the maturation of dendritic cells. The authors mentioned that hmwAPS presented more activities than low-molecular weight APS. Perera *et al.* (2018) showed that preconditioning of galactomannan from *A. cinnamomea* galactomannan (ACGM) enhanced immune response to invading bacteria early in infection but reduced the risk of severe inflammation later by reducing the secretion of pro-inflammatory cytokines. In addition, ACGM showed endotoxin-like effects on mouse macrophages. Table 8 lists the reported immune regulation activity of APSs.

Antioxidant activity of APSs

Oxidation is essential for numerous organisms to generate energy to drive biological processes. Normal energy metabolism in the brain necessitates the consumption of large amounts of oxygen by neurons to maintain biochemical activities in the body (Kawai *et al.*, 1989). According to a growing body of research, oxidative stress-induced cellular damage sets off the physiological processes of aging and most of the pathological developments that eventually result in significant health issues, such as Parkinson's and Alzheimer's diseases (Benzi and Moretti, 1995; Finkel and Holbrook, 2000).

Polysaccharides are the main natural antioxidant component of *A. cinnamomea*. Lin *et al.* (2010) indicated that in addition to the capability to scavenge 2,2-diphenyl-1-picrylhydrazyl free radicals, APSs had a strong antioxidant activity and thus could be used effectively as components of healthy or functional foods to alleviate oxidative stress response. Song and Yen (2002) showed the significant free radical-scavenging activity of polysaccharides from mycelia of *Antrodia cinnamomea* (PMAC). Tsai *et al.* (2007) also revealed that APSs in submerged fermentation exhibited a significant dose-dependent protective effect (of up to 200 μg/mL) on H_2O_2-induced DNA damage, which could alleviate H_2O_2-induced oxidative damage.

LPSs reduce the antioxidant and anti-inflammatory capacity, and damage the structure of cecal microflora of yellow-plumed chicken liver (Ye *et al.*, 2022). Ye *et al.* observed that the addition of APSs to feed improved the health conditions of chickens. This could be due to the combined effects of APSs on antioxidant and cytokine content and restoration of the declined beneficial flora of the cecum. As a result, the APSs improved the quality and yield of yellow feather chicken.

Production of APSs

As the wild fruiting body of *A. cinnamomea* is scarce and expensive, submerged fermentation has become the most efficient means to produce APSs (Li *et al.*, 2015). APS production can be dramatically improved by optimizing the fermentation conditions (such as temperature and pH) and medium composition (such as carbon, nitrogen, mineral sources, and vitamins) using a hybrid approach of artificial neural network and response surface model (Lin *et al.*, 2006, 2007). Sterol-type activators, including squalene, cholesterol, and stigmasterol, can increase APS production (Lin *et al.*, 2020). Squalene can greatly increase the contents of glucose, fucose, and mannose in APSs. Moreover, the addition of citrus peel extract benefits mycelial growth and AIPS production (Yang *et al.*, 2012). APS yield can also be increased by a two-stage pH fermentation process in shaker culture and stir-tank fermentation (Shu and Lung, 2004). During fermentation, the higher the oxygen supply, the more the APSs produced and the shorter the culture period (Shih *et al.*, 2006). In addition, the microparticle-enhanced cultivation technology can be used to increase the yield of bioactive compounds from *A. cinnamomea* in submerged fermentation (Fan *et al.*, 2023).

The residue of *A. cinnamomea* mycelium retains valuable active components after the extraction of polysaccharides or triterpenes. The dietary fiber extracted from the

Table 8. Immune regulation activity of APSs.

Sources	Immune regulation type	Optimum concentration	References
Mycelium	Cell-mediated immunity	-	Chen *et al.* (2008)
Mycelium	Cell-mediated immunity	10 μg/mL	Sheu *et al.* (2009)
Mycelium	Immunomodulatory effects on dendritic cells	200 μg/mL	Liu *et al.* (2010)
Mycelium	Activated dendritic cells	10 μg/mL	Lin *et al.* (2015)
Mycelium	CTX-induced immunosuppression	30 mg/kg	Liu *et al.* (2018)
Mycelium	Immunomodulatory effects on J774A.1 mouse macrophages, mouse peritoneal macrophages, and human dendritic cells	-	Perera *et al.* (2018)

residue of *A. cinnamomea* mycelium has a high purity and good adsorption capacity to oil, cholesterol, and sodium cholate *in vitro* (Xia *et al.*, 2022). In addition, using the *A. cinnamomea* mycelium residue as a feed additive in aquaculture significantly increased zebrafish feed efficiency and decreased fish inflammatory disease symptoms (Chang *et al.*, 2020). Thus, the reuse of *A. cinnamomea* residue provides a possible opportunity for the recycling economy of the *A. cinnamomea* industry to maximize utilization of *A. cinnamomea* and reduce economic losses.

Summary and the future perspectives

This review outlined recent research findings on APSs and summarized their extraction, isolation, composition, structure, and production. In addition, the biological activities, including anticancer, hepatoprotective, anti-inflammatory, neuroregulatory effects, antivascular, immune regulation, and antioxidant activity of APSs, were summarized. However, as a burgeoning resource, the development and application of *A. cinnamomea* is restricted by the following problems.

First, strengthening the industrialization of APSs requires breaking through the artificial cultivation technology of *A. cinnamomea*. At present, problems, such as long cultivation cycle, limited or insufficient source of raw materials, and poor fruiting body quality, are the bottlenecks restricting its industrialization development. Therefore, improving approaches, such as exploration of cultivation raw materials and innovation of cultivation methods, can alleviate the slow development of the industry.

Second, numerous products of APSs have been developed, but most of them are health products made from mycelium culture or crude extract of fermentation broth. Thus, the specific contents of the extracts are unclear, which is not conducive to the study and analysis of their pharmacological effects. In the future, further research on the separation and purification of monomers from *A. cinnamomea* could be carried out, which would lay foundation for the development of *A. cinnamomea* drugs.

In addition, the pharmacological effectiveness of APSs has been demonstrated at the molecular, cellular, and animal levels. However, a limited number of cases have reported their use in clinical adjuvant therapy or the treatment of major diseases. Therefore, the prospect of APSs in biomedicine can be further expanded, with a focus on the treatment or adjuvant therapy of major diseases.

Finally, throughout the *A. cinnamomea* research axis, major breakthroughs in germplasm resource protection and innovation, artificial cultivation, new compound mining, gene function, drug research, and development would be made in the future. Then, the standardization and marketization of the *A. cinnamomea* industry could be promoted globally to play an important role in the treatment and prevention of human diseases.

Author Contributions

Conceptualization: Hua-xiang Li and Lei Yuan. Methodology: Lei Yuan and Wen-yuan Zhou. Writing: original draft preparation, Jia-ning Dai and Bo-ling Liu; review and editing, Hua-xiang Li and Lei Yuan. Supervision: Dan Ji. Project administration: Hua-xiang Li and Dan Ji. Fund acquisition: Hua-xiang Li and Wen-yuan Zhou. All authors had read and agreed to the published version of the manuscript.

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Data are contained within the article and available upon request from the corresponding author.

Conflict of Interest

The authors declared no conflict of interest.

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