Pharmacological Actions of *Antrodia cinnamomea* and Its Cultivation Modes

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Abstract Being rich in various bioactive constituents, *Antrodia cinnamomea* is a kind of fungus having extremely high medicinal value. In this paper, the latest research progresses of pharmacological actions and cultivation modes of *A. cinnamomea* were reviewed.

Key words *Antrodia cinnamomea*; Pharmacological actions; Cultivation mode

1 Introduction

Antrodia cinnamomea, a kind of fungus belonging to Polyporales in Basidiomycetes, is rich in various bioactive constituents; the mycelium and fruiting body of *A. cinnamomea* are both edible. Having extremely high medicinal value, *A. cinnamomea* has the functions of anti-cancer, liver protection, anti-inflammation, anti-virus, anti-oxidation, adjusting immunity, anii-hyperlipemia and so on. Therefore, *A. cinnamomea* can nourish and build up the body, and support the healthy energy. Based on these, the latest research progresses of pharmacological actions and cultivation modes of *A. cinnamomea* were reviewed.

2 Pharmacological mechanisms of *A. cinnamomea*

2.1 Mechanism of anti-tumor High attention has been paid to *A. cinnamomea* due to its anti-tumor activity. Researches have shown that *A. cinnamomea* has good anti-tumor activity in both in vitro cell test and in vivo animal test. Kuo[1] *et al.* studied human hepatoma cell line Hep3B by the ethanol extracts of the fruiting bodies of *A. cinnamomea* (EAC). Results showed that EAC reduced the proliferation of Hep3B cells, so as to induced cell death. EAC enhanced the Ca^{2+} content in intercellular substances, and increased the expression of caspase-12 and calpain. Song[2] *et al.* researched on the effects of methanol extracts of fermentation mycelia of *A. cinnamomea* (MEM) on the human hepatoma cell line. Results showed that MEM blocked cell cycle and the caspase-3 and caspase-8 activation, restricted the proliferation of liver cancer cell and induced the hepatoma HepG2 cell apoptosis. Hseu[3] *et al.* studied on the apoptosis ability in vitro of promyelocytic leukemia HL-60 cell induced by *A. cinnamomea*. Results showed that *A. cinnamomea* restricted the proliferation and growth of HL-60 cell through inducing cell death. Liu[4] *et al.* researched on the effects of ethanol extracts of *A. cinnamomea* on the migration and proliferation of WEHI-3 cell in vitro, and explored the anti-tumor effects of EEAC in BALB/c mice engrafted with WEHI-3 cells. Results showed that EEAC reduced the proliferation and migration of WEHI-3 cells in vitro, decreased the chance of WEHI-3 cells entering into liver and spleen, and lowered the growth ability of tumor cells. Rao[5] *et al.* proved that Antrocin isolated from *A. cinnamomea* had restriction effects on metastatic breast cancer cell MDA-MB-231 (MMCs) through exerting effects on selective small molecule in Akt/mTOR signal pathway. Hseu[6] *et al.* argued that *A. cinnamomea* realized the inhibitory effects on melanoma cells through adjusting the Wnt/β catenin signal pathway.

2.2 Mechanism of liver protection *A. cinnamomea* has the functions of detoxication, anti-cancer and anti-inflammation, which is a traditional Chinese fungi drug for liver diseases having extremely high economic benefit. Song[7] *et al.* found out that fermentation broth of *A. cinnamomea* protected rat liver from acute oxidative damage induced by CCl_4; and preliminarily deduced that the liver protection ability was related to polysaccharides, polyphenols and triterpenoids. Lu[8] *et al.* researched the effects of ethanol extracts (Fr - I) of liquid fermentation medium of *A. cinnamomea* on the free radicals scavenging and acute liver injury induced by ethanol in rat liver. Results showed that the high- and low-dosage mycelium powders of *A. cinnamomea* could both avoid the increase of serum levels of aspartate transaminase and alanine aminotransferase induced by ethanol, maintained the MDA level of liver tissue, reduced the level of glutathione, aggregated the reduction of hepatic glutathione peroxidase (GPx) and glutathione reductase (GR) induced by ethanol. This indicated that the mycelium powders had significant effects of anti-lipid peroxidation; and its action mechanism was related to the increasing of liver GSH level and the GPx and GR activities of GSH related enzymes.

2.3 Mechanism of anti-inflammation So far, researchers have identified more than 20 chemical components from *A. cinnamomea* that having anti-inflammation effects, including maleic/succinic acid derivatives, triterpenoids, benzenoids, benzoquinone derivatives and polysaccharides. Hseu[9] *et al.* took macrophage RAW264.7 as the research materials, and observed the in vitro anti-inflammatory effects of *A. cinnamomea* mycelium culti-

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vated by deep liquid fermentation method. Results showed that *A. cinnamomea mycelium showed no significant cytotoxicity at the* concentration of 0 – 50 μg/mL, but could significant restrict the production of cytokine NO and PGE2 in RAW264.7 cell induced by lipopolysaccharide LPS and reduced their expression levels. Further researches found out that these changes were casued by the reduction of iNOS and COX2 protein expression expression level in cells. iNOS and COX2 were key proteins for NF-κB anti-inflammatory pathway; reduction of their expression levels indicated that *A. cinnamomea* mycelium exerted the in vitro anti-inflammatory effects through NF-κB pathway.

Chen[10] *et al*. illuminated the anti-inflammatory mechanism of *A. cinnamomea*. Researchers found out the five triterpenoids antcin A, B, C, H, K isolated and purified from the fruiting bodies of *A. cinnamomea*, and argued that antcin A in these five chemical components had most similar chemical structure like cortisone and dexamethasone. Therefore, antcin A led to the transfer of glucocorticoid receptors (GR) in human lung adenocarcinoma A549 cell from cytoplasm to cell nucleus. Due to the lipophicity, antcin A was easily diffused through cell membrane, and combined with the glucocorticoid receptors (GR) in cytoplasm. After the combination, GR separated from heat shock protein; bipolymer was formed by GR/antcin A compound and finally transferred into cell nucleus. Then, GR/antcin A was combined with glucocorticoid responsive elements (GRE) of target gene, so as to regulate the expression of gene, such as restricting the expression of proinflammatory protein, and enhancing the expression of anti-inflammatory protein.

2.4 Mechanism of immunomodulation Modern researches have found out that some Chinese herbal medicines have bidirectional way regulating immune functions, so as to make high or low immunoreaction return to normal. The material basis of immunomodulator contains polysaccharides, glycosides, terpenes, flavonoids, volatile oils and so on. Meng Fan-yue[11] *et al*. inoculated ascites of H22 tumor strain into subcutaneous tissue of the back of mice in order to establish tumor bearing model. Mice were fed with fruiting body powders of *A. cinnamomea* at low-dosage [40(BW) mg/kg], middle-dosage [200(BW) mg/kg] and high-dosage [1 000(BW) mg/kg] groups, respectively. Results showed that fruiting bodies of *A. cinnamomea* enhanced the anti-tumor immune function of tumor bearing mice at the levels of immune cells and immune molecules, showing a dosage dependent trend. Therefore, the antitumor effects of fruiting bodies of *A. cinnamomea* were related to the functions of enhancing immune system and increasing anti-tumor ability.

2.5 Mechanisms of other functions Many researches have proved that except the functions mentioned above, *A. cinnamomea* also had effects of antioxidation, neuroprotection, antiplatelet aggregation, anti-hypertension, anti-hyperlipemia and so on. Hseu[12] *et al*. researched on the inhibitory effects of water extracts from *A. cinnamomea* mycelium on the human erythrocyte hemolysis and lipid /protein peroxidation. AAPH is a kind of water-soluble free radical inducer, which stimulates the oxidation reaction in organism and the generation of hydrogen peroxide radicals, and causes a series of lesion due to the damage of erythrocyte by free radicals. Water extracts from *A. cinnamomea* mycelium restrict the free radicals damage induced by AAPH, effectively weaken the oxidative hemolysis and lipid peroxidation of erythrocyte, and protects the cytoplasmic antioxidant glutathione. Cheng[13] *et al*. found out that *A. cinnamomea* polysaccharides restricted the vascular endothelial growth factor VEGF, blocked the formation of capillary blood vessels and the migration of endothelial cells induced by VEGF, inhibited the expression of cyclin D1 and the generation of neovascularization. Li[14] *et al*. found out that *A. cinnamomea* reduced the formation of new blood vessels in rat carotid artery, effectively restricted the proliferation and migration of aortic smooth muscle cell induced by PDGF, and reduced the neointima formation caused by external carotid artery occlusion. This indicated that *A. cinnamomea* had good effects on curing dyslipidemia and atherosclerosis.

3 Cultivation methods of *A. cinnamomea*

The results of pharmacological research have promoted the application of *A. cinnamomea*. Due to the limited resources, cultivation technology of *A. cinnamomea* has become another research focus. At present, the cultivated *A. cinnamomea* mycelium and fruiting body have been mature in technology. And the cultivated *A. cinnamomea* has similar content of total triterpenes like the wild plant. Shenzhen *A. cinnamomea* Laboratory of Hong Kong Chinese Medical Science Academy developed many cultivation technologies of *A. cinnamomea*, such as submerge fermentation of *A. cinnamomea* containing high triterpene. The artificial cultivation technology of *A. cinnamomea* has achieved great breakthrough, which lays solid foundation for the development of *A. cinnamomea*.

3.1 Infected cultivation and artificial basswood cultivation Cultivated fruit body is obtained, the active components of which are close to the wild *A. cinnamomea*. However, few basswoods could be found, the fruiting body grows slowly with high production cost. Besides, fruiting bodies are easily affected by aflatoxin, heavy metal and other harmful ingredients. Therefore, the quality of the fruiting bodies of *A. cinnamomea* is hard to be controlled; and the standardized and large-scale production of *A. cinnamomea* could hardly be achieved.

3.2 Solid state fermentation This method is also called space bag fermentation. It significantly reduces the production cost, shortens the production cycle to about three months, and realizes automatization and industrial production in a certain degree. The fermentation products are *A. cinnamomea* mycelium; the content of total triterpene reaches 3% – 5%. The crude extracts of fermentation products have almost the same effects as that of wild *A. cinnamomea*. However, production cycle of three months still can not greatly reduce the production cost; large-scale production of *A. cinnamomea* could hardly be achieved; the quality of the fruiting bodies of *A. cinnamomea* is hard to be controlled; and the

quality management is difficult.

3.3 Submerged liquid fermentation Submerged liquid fermentation is a necessary method for the industrial production of most dical fungi, which has the advantages of high degree of automation, short production cycle, low production cost, and controllable quality. Jiangnan University [15-16], Hong Kong Chinese Medical Science Academy [17-18] and other research institutes and biotechnology companies have devoted considerable resources to study the liquid fermentation products of *A. cinnamomea*. At present, the total triterpene content in *A. cinnamomea* by the method of submerged liquid fermentation reaches as high as 11.9%, which is higher than those by solid state fermentation and dish cultivation. Triterpene is the key pharmacological ingredient of *A. cinnamomea*, as well as an important index to evaluate the quality of *A. cinnamomea*. Achieving high content of triterpene provides foundation for the in-depth research of *A. cinnamomea*, especially the production of anti-cancer drugs and anti-hepatic injury drugs based on *A. cinnamomea*. This research results will greatly promote the industrialization of *A. cinnamomea* industry.

4 Analysis of research status and its prospect

As a rare medicinal fungus in Taiwan, *A. cinnamomea* has been paid high attention for only 20 years since it is found in the 1990s. Researches have been transferred from the identification of biological function of total extracts to the identification of a specific function target of single active component [19]. Many animal model tests are introduced into the identification of different components of *A. cinnamomea* and their activities, such as anti-tumor, liver protection, anti-inflammation, immunomodulation and antioxidation. Pharmacological mechanism of *A. cinnamomea* in tests in vivo can be applied in the clinical research in future.

Due to the rare resources, market price of *A. cinnamomea* is very high. In order to protect this precious resource and to meet the increasing demands of people, more attentions are paid to the large-scale cultivation technology so as to obtain triterpene and other effective components, as well as on the synthesis method to obtain biological active component of *A. cinnamomea*.

A. cinnamomea can improve the health status of human to a great degree. However, *A. cinnamomea* is only used as a food supplement not drug in the market. It is hoped that by using the clinical test method, *A. cinnamomea* or its components can be transformed into drugs, so that this rare and precious fungus can be fully used.

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