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Structure, bioactivities and applications of the polysaccharides from Tricholoma Matsutake: a review

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Abstract
Tricholoma Matsutake is an important edible mushroom that has been artificially cultivated and used as a food and medicinal ingredient in traditional Chinese medicine. In the past decades, many researchers have reported that Tricholoma Matsutake Polysaccharides (TMP) possess various bioactivities, including anti-tumor, immunomodulatory, anti-oxidation and anti-aging. The structural characteristics of TMP were studied by GC-MS, LC-MS, NMR, and FT-IR, and the structure-activity relationship of TMP biomolecular was analyzed. This paper reviews the progress of TMP extraction, purification, structure characterization and application.

Keywords: Tricholoma Matsutake polysaccharide; structure; bioactivities; applications.

Practical Application: The practical application is to provide a comprehensive source of information for researchers and consumers, so they can better understand Matsutake polysaccharides and their biological activities to carry out more clinical studies to meet the criteria for new drug development, and more convincing scientific data should be provided. Matsutake polysaccharides can be better recognized and applied all over the world.

1 Introduction

Matsutake belongs to the order Agaricales and the family Tricholoma Mataoeae,which has many common names, such as agaric. It is a rare edible fungus and is mainly distributed in Japan, the Korean Peninsula, Northeast China, Yunnan, Taiwan, and other regions. Matsutake is a valuable wild edible and medicinal fungus with thick flesh and delicious taste and it is rich in protein, polysaccharides, amino acids, and dietary fiber. It is considered as one of nature's popular food and most precious herbal medicine ingredient and known as the "king of fungi". Recent studies reported that Tricholoma Matsutake polysaccharides (TMP) are viewed as ideal ingredients for healthy food and pharmaceutics since it has many bioactivities such as improving immune function, anti-cancer, anti-aging, and anti-radiation. While there are many research studies on mushrooms, there are few reviews on TMP. This article reviews the extraction, isolation, purification, structural characterization, biological activities, and applications of TMP (Figure 1).

2 Extraction and purification

In the existing research, most of studies have focused on the extraction of polysaccharides from the fruiting bodies and mycelium (Wang et al., 2019). Extraction is a crucial and indispensable step for obtaining polysaccharides and can affect polysaccharide yield, quality, chemical structure, and even biological activities (Gong et al., 2020). Temperature, reagent type, and extraction time all affect extraction efficiency and biological activity (Wang et al., 2021a). Up to now, several conventional extraction techniques have been used to extract TMP like hot water extraction, ultrasonic extraction, microwave extraction, alkaline extraction, microwave-assisted extraction, ultrasonic-assisted extraction, and compound enzymatic hydrolysis extraction (Leong et al., 2021; Barbosa et al., 2020).

2.1 Hot Water Extraction (HWE)

The usually extracted by stirring the crushed fruiting bodies in hot water for several hours. This extraction method is simple to carry out, but takes a long time, has a large amount of solvents, and a high temperatures (Barbosa & Carvalho, 2020). Response surface optimization was used to improve extraction rate and yield of polysaccharide (Wang et al., 2021b; Yuan et al., 2020).

2.2 Ultrasonic Extraction (UAE)

The UAE uses cavitation to release great amount of energy, generating shock waves, microjets, and high shear force (Zheng et al., 2021). Hydrodynamic can promote cell wall disruption, immiscible phase mass transfer, improve penetration and capillary effects as well as decrease particle size, thus increase yield and efficiency (Zhao et al., 2013). Compared with the conventional hot water extraction method, UAE can raise the extraction yield and purity, save operation time and streamline the operation process. The study showed that ultrasonic power

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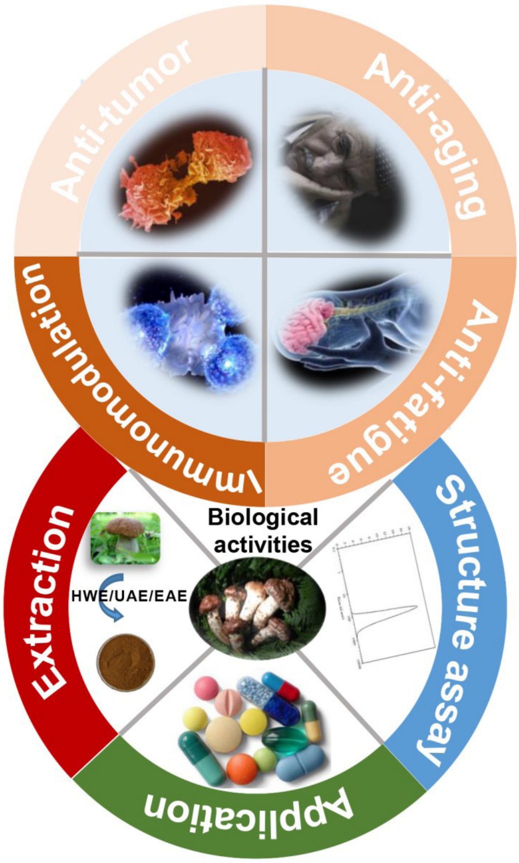

Figure 1. Overall schematic representation of polysaccharides from Tricholoma Matsutake.

and frequency, liquid-solid ratio, extraction temperature, and time had significant effects on the extraction of polysaccharide (Zhao et al., 2018). Compared with the traditional hot water extraction method, the method of applying ultrasonication-assisted hot water extraction method has the advantages of high polysaccharide extraction rate, minor-cycle, and low cost, and has important practical significance for the industrialized large-scale production of polysaccharides (Cui et al., 2018).

2.3 Microwave Extraction (MAE)

Microwave is a kind of non-contact heat source, which produces heat energy through ionic conduction between solvent and dissolved ions, so as to break cell walls and accelerate the release of polysaccharide molecules (Delazar et al., 2012). MAE is advantageous with improved efficiency, reduced extraction duration, rapid and volumetric heating of the absorbing medium, less solvent consumption, better selectivity of target molecules, and potential for automation for polysaccharides extraction from mushrooms (Marzuki et al., 2021). A study has optimized MAE of TMP conditions for operating extraction of polysaccharides which optimal conditions obtained 66 w, 1 : 25, and 45 min (Yao et al., 2008). A large number of studies have shown that MAE and UAE have the advantages of high extraction rate, low solvent consumption, flexible operation equipment and short extraction time compared with traditional extraction methods. However, the main disadvantages of microwave-assisted extraction were that the microwave power was too high and the heating was not inhomogeneous, which reduced the extraction rate of polysaccharide (Zeng et al., 2012). Therefore, instrument power, extraction temperature, and time are significant parameters to be considered polysaccharides extraction (Zhu et al., 2016a). A study by Yin and colleagues suggested that hot water-assisted with ultrasonic technology to extract Matsutake polysaccharides which were optimal conditions obtained 95 °C, 1 : 12, and 3.5h.

2.4 Enzyme-Assisted Extraction (EAE)

Enzymes can effectively catalyze hydrolysis and degradation of fungal cell wall matrix, resulting in better release and more efficient extraction of bioactive compounds within cells (Marić et al., 2018). EAE has the advantages of simple operation, high specificity, environmental-friendliness, high efficiency, low energy consumption, and usually does not destory the molecular structure of polysaccharides (Veeraperumal et al., 2021). Nevertheless, the relatively strict temperature and PH of extraction are some of the affected factors of EAE. Cellulase, papain, trypsin, and pectinase-assisted extraction contributed to breaking down the physiochemical linkages between protein and other molecules and assist in the release of TMP leading to enhanced extraction yield (Yin et al., 2014).

2.5 Alkaline or Acid Extraction (AE)

During the extraction process, alkaline or acid extraction was used to facilitate the release of mushroom polysaccharides during extraction. Acid and alkaline treatments destroy cell walls and degrade the crude fiber structure, as well as hydrolyzable linkages between cell wall protein and glucan (such as linkages with O-linked side chain), allowing the release of intracellular polysaccharides, extracting soluble fraction of acid or base, and also converting insoluble components into water-soluble one (Zhu et al., 2016b). Acid-extracted polysaccharides have a low fraction compared with alkaline extraction which might be due to the disruption of hydrogen bonding (Jing et al., 2018). Study reported that the extraction rate of polysaccharides by mild alkaline hydrolysis from matsutake was significantly higher compared with hot water extraction. MAE and alkaline extraction had a prominently higher yield, suggesting that approximately 80% of the water-soluble polysaccharides were extracted (Wang et al., 2019).

Among steps of studying fungus polysaccharides, extraction is the most significant stage as it determines the monosaccharide composition, molecular weight, structure, spatial configuration, and bioactivity. Since most functional polysaccharides in mushrooms are water-soluble, they can be assisted with water extraction using physical techniques such as heating, ultrasound, microwave, and others. Comparing different extraction techniques, MAE and UAE generally required the shortest time. For the case of operating temperature, HWE usually operated at elevated temperature has the highest operating temperature. On the other hand, HWE has the highest energy consumption due to its long extraction time and high operating temperature, while UAE has the lowest energy consumption (Table1). In some extraction

Table 1. Advantages and disadvantages of TMP extraction methods.

Extraction method	Advantage	Disadvantage
HWE	Easy to carry out	Long extraction time
		Large volumes of solvents Elevated temperature
UAE	Higher purity	Destroy structure
	Lower energy consumption	
	Shorter operating time	
MAE	Shorter operating time	Inhomogeneous heating
	Higher extraction efficiency	
EAE	Easy to carry out	Strict temperature and PH
	Simple equipment	
	Usually don't damage the TMP molecular structure	
Alkaline or acid extraction	Easy to carry out	Affect structure and biological activity
HWE + UAE	High extraction efficiency	
	Short cycle	
	Lower cost	
Alkaline + MAE	High extraction efficiency	
	Shorter operating time	

of polysaccharides, EAE achieved a significantly higher yield compared with HWE and UAE for polysaccharides. In recent years, the osmotic pressure method, high-voltage pulsed electric field method, supercritical CO_2 extraction method, and two-phase extraction method have also been applied to the extraction of edible fungus polysaccharides (Ren et al., 2019).

2.6 Purification

The general procedures for the separation and purification matsutake polysaccharides were as follows: pretreatment of the total fruiting bodies or mycelium of matsutake with ethanol under reflux to remove oil. The remaining residue was extracted by drying. The solution was collected after filtration and concentration (Ren et al., 2019). Crude polysaccharides were obtained by alcohol-precipitation, dissolved in water and insoluble residues were removed by centrifugation. Different column chromatography including anion exchange chromatography such as DEAE-Sepharose, DEAE-Cellulose, and DEAE-Sephadex, as well as gel permeation chromatography (Sephadex G, Sephacryl S, Sepharose CL) were used to obtain purified polysaccharides (Du et al., 2013). Ding and colleagues used the Sevag method to deproteinize, put crude polysaccharides on a DEAE cellulose column, eluted with stepwise NaCl gradient, and then concentrated, lyophilized, and purified the eluate with a Sephadexg-100 column. The TMP obtained was named TMP-A, and the yield was 0.22% (Ding et al., 2010). TM-P5, a new water-soluble polysaccharide fraction, was used water decoction-alcohol precipitation extraction method and purified the crude polysaccharide into 9 components (TM-P1-9) by a sephacryls-300 column. The purified is further applied to the sephacryls-300 column for obtaining TM-P5 (You et al., 2013).

3 Assay for polysaccharides

Polysaccharides were a kind of long and complex chains carbohydrates and involved in biological processes, such as embryonic development and cellular immunity to infection by viruses or bacteria (Li et al., 2016). Mushrooms contain many types of polysaccharides, which include heteropolysaccharides rich in galactose, fucose, mannose, and xylose. Since TMP was reported by Takusaburo Ebina in 2003 (Ebina, 2003). More than ten polysaccharides were obtained from the fermentative mycelia and fruiting bodies of Tricholoma matsutake.

Their structural characteristics were investigated by Fourier transform infrared spectroscopy (FT-IR), nuclear magnetic resonance (NMR), gas chromatography-mass spectrography (GC-MS), methylation analysis, periodate oxidation-Smith degradation, partial acid-hydrolysis, and enzymatic degradation. High-performance liquid chromatography (HPLC) could be used to determine of molecular weight of purified polysaccharides. Ultraviolet Spectroscopy (UV), Infrared Spectroscopy (IR), Gas chromatography (GC), Mass Spectrometry (MS), and Nuclear Magnetic Resonance (NMR) can be determined the structure of the polysaccharide (Jia et al., 2004). Periodic acid oxidation, Smith degradation, reductive cleavage, and acetylation methods were commonly be used to determine the types of glycosyl bonds (Wu et al., 2015). The placement and sequence of monosaccharide residues and the position and chirality of glycosidic residues result in the potential high structural variability of polysaccharides and affect polysaccharide bioactivities (Ren et al., 2019). As a result, it is of great significance to determine the polysaccharide structure. TMP are usually partially acid hydrolyzed into oligosaccharide fragments, and then the main components are determined by methylation or GC analysis composition of chain and branch. Here in, we have listed the reported TMP over the past decade and have provided comprehensive information on their molecular weight, monosaccharide composition, and associated references, as shown in Table 2.

TMP-5II was isolated from the fresh fruiting bodies of Tricholoma matsutake. Monosaccharide compositions were D-glucose, D-galactose, D-mannose, and D-fucose. IR and NMR spectra showed that TMP-5II had (1-4)-beta-pyran glucose as the main chain, and a branched-chain in the O-6 location with

Table 2. The polysaccharides isolated from Tricholoma Matsutake.

No	Name	Monosaccharide composition	M.W. (Da)	Reference
1	TMP5II	D-Glu, D-Gal, D-Man, and D-Fuc.	1.58×10^4	Cheng et al., 2016
2	TMP-B	α-D-Glu and α-D-Gal in a ratio of 7:2.	1.64×10^4	Hou et al., 2017
3	TMP-A	D-Glu, D- Gal, and D- Xyl in a ratio of 79.37:9.81:10.82.	8.89×10^4	Ding et al., 2010
4	TMP30	L-Fuc, D-Gal, D-Glc, D-Xyl, and D-Man in a ratio of 9.3:26.8:40.1:2.6:16.4.	Unknown	Chen et al., 2017
5	TMP60	L-Fuc, D-Gal, D-Glc, D-Xyl, and D-Man in a ratio of 6.6:17.6:42.3:12.1:21.1.	Unknown	Chen et al., 2017
		L-Fuc, D-Gal, D-Glc, D-Xyl, and D-Man in a ratio of 8.1:21.2:43.0: 4.2:23.6.		
6	TMP80		Unknown	Chen et al., 2017
7	TM-P1	Glu, Gal, and mannose in a ratio of 5.9:1.1:1.0.	3.61×10^5	You et al., 2013
8	TM-P2	Glu, Gal, Man, Fuc in a ratio of 7.7:4.0:1.9:1.0.	3.72×10^5	You et al., 2013
9	TM-P3	Glu, Gal, Man, Fuc in a ratio of 5.1:4.9:2.2:1.0.	3.65×10^5	You et al., 2013
		Glu and Glucuronic acid.		
10	TM-APS-1	Glu and Glucuronic acid.	2.8×10^5	Tong et al., 2013
11	TM-APS-2		5.4×10^4	Tong et al., 2013

fucose (1-2) mannose (1-3)-alpha-pyran galactose (Cheng et al., 2016). TMP-B isolated from fruiting bodies of Tricholoma matsutake were composed of α-D-glucose and α-D-galactose which ratios were 7 : 2 and had a backbone of 1, 4-linked α-D-glucose which branches were mainly composed of two 6-linked α-D-galactose residues, and the α-D-galactose were 1, 6-linked (Hou et al., 2017). IR, GC-MS, and NMR spectra showed that TMP-A had a backbone of 1,4-β-d-glucopyranose residue which branches at O-6. The branches were mainly composed of a (1→3)-α-d galactopyranose residue, and terminated with α-d-xylopyranose residue (Ding et al., 2010). Three novel TMP fractions (TMP30, TMP60, and TMP80) were isolated and purified from TMP by stepwise alcohol precipitation. Their preliminary structural features were determined by high-performance anion-exchange chromatography with HPAEC-PAD and FT-IR analyses. The results suggest that pyranoses existed in the β-configuration in TMP30, TMP60, and TMP80 (Chen et al., 2017). Three polysaccharides (TM-P1, TM-P2, TM-P3) were isolated from the fresh fruiting bodies of Tricholoma matsutake. IR and NMR spectra showed that TM-P1 were mainly composed of glucan and galactan and backbone chains were composed of 2,3,6-linked glucose. TM-P2 was mainly composed of a Glcp backbone with mostly 1,6-links and 1,4-links. TM-P3 was composed mainly of glucan and galactan, which was similar to that of the TM-P2 (Cheng et al., 2016). However, TMP-A was determined that 1,4-linked glucose represented the largest amount of residues of the TMP structure and the branched residues were 1,4,6-linked glucose. These discrepancies may due to the differences in polysaccharide species and extraction methods. TM-APS-1 and TM-APS-2 were polysaccharide fractions isolated from the fruit bodies of Tricholoma matsutake and composed of glucose and glucuronic acid (Tong et al., 2013).

4 Bioactivities

As different structural characteristics and widely biological activities, TMP act as a "biological response modifier" and have been considered a promising bioactive compound that exhibits a range of biological activities: antioxidant, anticancer, antitumor, anti-inflammatory, anti-coagulation, anti-radiation, anti-fatigue, and immune-modulating activities (Figure 2).

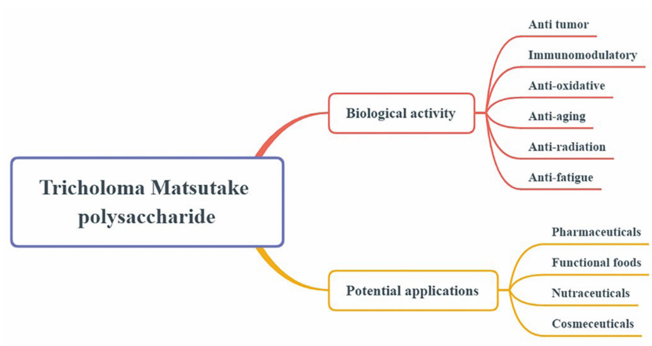

Figure 2. The biological activity and industrial application prospects of TMP.

4.1 Anti-tumor

The World Health Organization has reported that 8.2 million people die of cancer each year, accounting for 13% of all death worldwide. In recent years, studies on the mechanism of polysaccharides have shown that fungus polysaccharides inhibit tumor growth and reduce side effects during treatment of tumors, and can enable patients to rebuild their immune system improving their cancer resistance. For instance, Tuckahoe polysaccharide exerts anti-tumor activity by killing the host's tumor cell (Li et al., 2019). Early in 1983, researchers have reported that polysaccharides isolated from the cultivated fruiting body of matsutake showed high antitumor activity. Another recent study demonstrated that TMP has high anti-tumor activity in mice transplanted with S180 sarcoma cells. These fungus polysaccharides couldn't only prolong the longevity but significantly reduce the mortality of the hosts. Polysaccharides mainly play an immune function through macrophages, and macrophages can respond to infections, tumors, and inflammation. Macrophages directly kill pathogens through phagocytosis and present antigens to elicit an immune response. Macrophages produce a large number of biologically active molecules, including nitric oxide (NO), reactive oxygen species (ROS), and cytokines including tumor necrosis factor TNF-α, IL-1α, and IL-1β, for the defense of IL-6, IL-10 (Christopher et al., 2021; Yin et al., 2019). TMP can promote mice's lymphocytes and macrophages (Hou et al., 2017; Hou et al., 2013). Ding performed a proliferation assay, phagocytosis assay, and cell cycle analysis of RAW264.7 macrophages to identify differentially signaling expressed passway and found the molecular mechanisms associated with differences in the anti-tumor activity of TMP in macrophages and significantly anti-tumor activity is expressed through MAPK and NF-κB signaling pathways, ultimately (Ding et al., 2016; Li et al., 2021).

The anti-tumor activity of TMP is closely related to its concentration, and its inhibitory effect on tumor cells shows time and concentration-dependent (Hou et al., 2013). The low concentration of TMP has little effect on HepG-2 and HS766T cells, but the inhibitory effect on Tca8113, Hela, and MCF-7 cells is obvious and the inhibitory effect shows a certain dose-effect relationship and time-dependent (Liu et al., 2013). The study established in vitro models of TMP-2 component antioxidant and anti-tumor activity, and the present results suggested that when TMP-2 was 4.0 mg/ml, it was effective against HepG2 and A549 cells (You et al., 2013) (Figure 3).

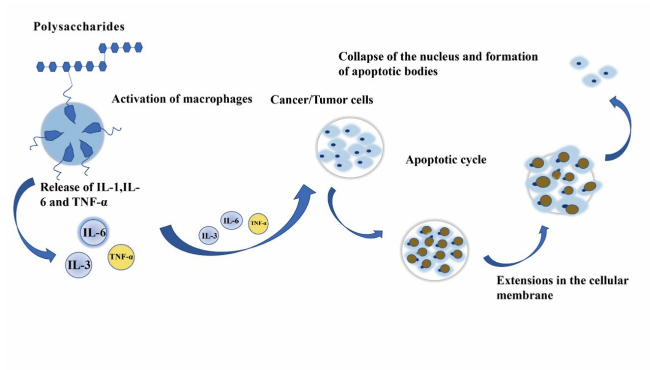

Figure 3. Effect of polysaccharides *via* apoptosis induction of tumor cells.

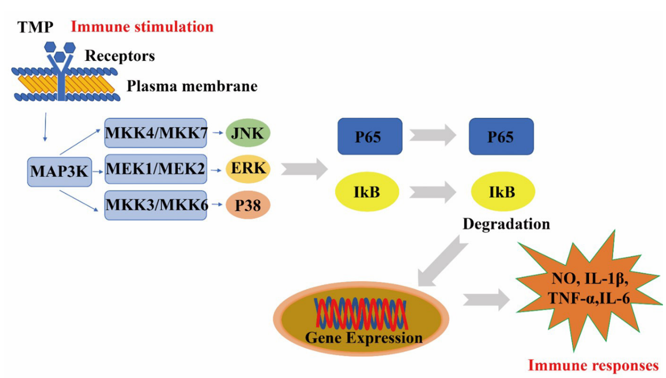

Figure 4. Immunomodulatory mechanism of TMP in macrophage.

4.2 Immunomodulation

Immune activity can modulate the body's immune functions by regulating immune organs, immune cells, immune molecules, and their immune activity without significant side effects. Polysaccharides mainly play an immune function through macrophages, and macrophages can respond to inflammation and infections. Macrophages directly kill pathogens through phagocytosis and present antigens to elicit an immune response (Byeon et al., 2009). Aging results in a significant decline in immune function (immunosenescence), such as reduced proliferation of circulating T-cells, increased production of pro-inflammatory IL-6, IL-1β, TNF-α, and diminished NK cell activities (Ren et al., 2021). Byeon studied the immunomodulatory effect of TMP by using a functional activation model of macrophages, monocytes, and splenic lymphocytes and he found that TMP can augment the level of NO and TNF-α (Byeon et al., 2009). TMP can inhibit modulate cyclophosphamide-induced mouse leukopenia and regulate NK cells and lymphocytes. Therefore, it is concluded that TMP can effectively improve the immune function of immune-suppressed mice by regulating ILs and inflammatory factors related to the NF-κB signaling pathway (Li et al., 2017) (Figure 4). The immunomodulatory activity of TMP can be observed by studying the activity of interferon, the content of enzymes related to immune regulation, and increasing the content of immune cells. Different solvents extraction of TMP on indicators such as thymus index, spleen index, and macrophage phagocytic rate were scientifically measured and found TMP of water extract (WE) and n-butanol extract (BAE) can significantly improve the immune indicators of mice and TMP can promote macrophage proliferation, promote cytokine release and gene expression by affecting G0/G1, S and G2/M phases (Yin et al., 2012).

4.3 Anti-oxidation

TMP has anti-oxidant activity, reducing hydroxyl radicals and superoxide radicals, and enhancing reducing power (You et al., 2013). Hydrogen peroxide induces oxidative stress and apoptosis of skin fibroblasts in a concentration-dependent manner, and decreases human skin fibroblasts, while increasing ROS generation and cell apoptosis (Agrawal et al., 2021). TMP pretreatment reduces oxidative stress, cell apoptosis in hydrogen peroxide-treated skin fibroblasts, and decreases the levels of superoxide anion free radicals, DPPH free radicals, and hydroxyl free radicals, and can significantly reduce the damage caused by hydrogen peroxide to PC12 cells to achieve anti-oxidant and anti-aging effects (Ding et al., 2010; Tong et al., 2013) (Figure 5).

The solvent is also a factor that affects its antioxidant activity. The TMP was extracted with petroleum ether, chloroform, ethanol, and water at their corresponding solvent boiling temperatures. Ethanol extracted part was extracted with ethyl acetate and n-butanol in turn and the antioxidant activity of each extract. The results showed that diverse solvent extracts of matsutake certain antioxidant activity, of which ethyl acetate and water extract have strong antioxidant activity (Yin et al., 2011). The antioxidant capacity of different components of TMP is also discrepant. Gao Qing and colleagues performed oxygen

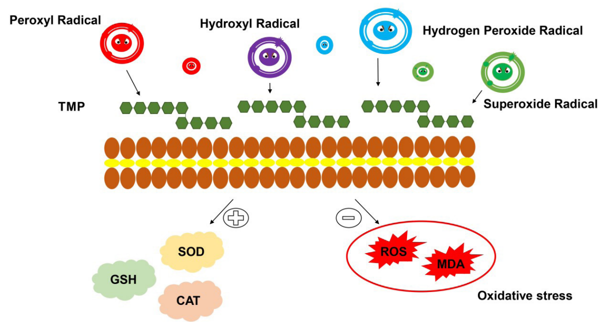

Figure 5. Anti-oxidation mechanism of TMP.

scavenging free radical determination on the extracted TMP, which showed that had high oxygen-free radical scavenging ability. It has been the antioxidant activity of TMP was evaluated in a study using multiple free-radical scavenging methods, wherein they determined the scavenging abilities of oxygen free radical of four components TMP-1A, TMP-1B, TMP-2A, and TMP-2B. They found that TMP-2 has the strongest in vitro antioxidant activity (You et al., 2013). Ultrasonic-assisted extraction technology was used to extract crude polysaccharides from the fruiting bodies of Matsutake. By measuring the scavenging rate of 2,2-diphenyl-1-picric acid (DPPH) and hydroxyl free radicals in vitro antioxidant activities, the results showed the order of the antioxidant activity of each sub-fraction of TMP is TMP80 > TMP > TMP60 > TMP30. These findings indicate that TMP80 may be a promising bioactive macromolecule (Chen et al., 2017).

4.4 Anti-aging

As the worldwide population ages, increasing attention is being paid to the physical and health needs of the population. The dream of fighting aging is as old as human civilization, so interventions to care for aging and delay age-related diseases are essential (Carmona-Gutierrez et al., 2019). Aging is generally defined as the cumulative changes in various pathologies that increase the risk of disease and death (Wang et al., 2017). Aging is a process in which the functions of various tissues and organs of the body undergo degenerative changes with age. It is closely related to diseases such as hypertension, diabetes, atherosclerosis, and Alzheimer's disease. Aging is an inevitable law of the life process. The research on the mechanism of aging to find effective anti-aging drugs has become a hot-button issue in the field of aging research. Establishing animal models similar to clinical aging symptoms is an effective means to study aging mechanisms and evaluate anti-aging functions. Traditionally, chemical analysis methods and several in vitro and in vivo detection methods involving various cell types and animal species have been used to identify the anti-aging activity of edible fungi, but none of these methods have proven to be sufficiently sensitive. Anti-aging in vivo is mainly studied by comparing the activity and content of enzymes in blood, liver, spleen, heart, and kidney between model and control mice. Studies have shown that TMP has anti-aging activity, and the concentration of polysaccharides is positively correlated with its activity. Polysaccharides containing selenium and zinc increase the ability to release free radicals, and they are a potential source of natural antioxidants and anti-aging substances. TMP can scavenge free radicals, inhibit or block lipid peroxidation caused by free radicals, increase the activities of SOD, CAT, and GSH-Px, improve the body's antioxidant capacity, spleen index, spleen cell ConA's proliferation response, macrophage phagocytic function, antibody production ability, and achieve anti-aging effects (Zhu et al., 2020).

4.5 Anti-radiation

Ultraviolet rays, mobile phones, computers, some high-frequency electrical appliances, and hospital radiation equipment are all sources of radiation that cause adverse effects, so that radiation protection is getting more and more attention. Ultraviolet radiation can also lead to excessive production of free radicals especially. Excessive ROS, including superoxide anion (·O2-), hydroxyl radical (·OH), and hydrogen peroxide (H_2O_2), disrupt the homeostasis of the antioxidant defense systems in the epidermis, causing oxidative stress, leading to DNA damage, activing and changing cell or tissue growth, differentiation, senescence, and connective alterations (Wang et al., 2020). TMP has a significant protective effect on the hematopoietic function reducing the radiation sensitivity of hematopoietic stem cells and hematopoietic stromal cells, shortening cell cycle disorders, and promoting the recovery of the body's hematopoietic function (Wang, 2008).

4.6 Whitening activity

The role of whitening agents inhibits the activity of tyrosinase or block the oxidation pathway of tyrosine to produce melanin, thereby reducing the production of melanin to achieve skin whitening effect (Du et al., 2014). Cheng Hua and colleagues studied TMP inhibited tyrosinase activity well with an IC50 value of 136.4 mg/L (Cheng et al., 2013). The polysaccharide of Matsutake fungus silk has a prominent inhibitory effect tyrosinase activity, which is much higher than other polysaccharide components, and clearance rates of DPPH· and ·OH are as high as 90.06% and 31.43% respectively. It is in vitro for thiobarbital lipid peroxidation inhibition rate is 87.7%, IC50 for tyrosine monophenolase and diphenolase are 1.18 μg/mL, 1.46 μg/mL respectively, which is a reversible mixed inhibition, which is developed into a whitening product for mycelial polysaccharide provides basic theory (Wang, 2015). TMP has scavenged DPPH free radicals, inhibited tyrosine activity and whitening effect, the development of TMP into whitening cosmetics has good application prospects.

4.7 Anti-fatigue

Studies have reported that two weeks of treatment with matsutake can improve the exercise performance of mice in load-bearing swimming, rotating rod, and forced running tests, including levels of adenosine triphosphate (ATP), antioxidant enzymes, and glycogen in muscle, liver, or serum increase, the level of malondialdehyde and active oxygen decrease (Li et al., 2015). The research showed that both TMP has anti-fatigue effects, and the finding in the research can be used as a valuable resource for further identification and provide experimental evidence for clinical trials of Matsutake as an effective drug for anti-fatigue related diseases.

4.8 Other

Hypertension is the main risk factor for cardiovascular disease. Studies have found that TMP at a dose of 400 μmg/kg has a hypotensive effect on spontaneously hypertensive rats (SHRs) (Geng et al., 2016). Therefore, Matsutake can be used as a functional food to help to prevent hypertension-related diseases.

5 Conclusions and prospects

Matsutake, a rare plant fungus, has excellent nutritional value and also remarkable potential applications for medicinal purposes. Nutritionally, Matsutake is a good source of carbohydrates, proteins, amino acids, micronutrients, and dietary fiber. Matsutake has been demonstrated to possess a wide variety of bioactivities, particularly anticancer, immunomodulatory, anti-oxidation, and anti-aging effects (Xu et al., 2015). However, the exact mode of action of these carbohydrates is still elusive and deserves special attention in the future study (Barbosa et al., 2020). Extensive research has also focused on TMP, TMP isolated from Matsutake, which exhibits both promising anticancer and immunomodulatory effects. The majority of the studies showed congruent findings that the TMP elicits its immunomodulatory effects by MAPK and NF-κB immune passways. Apart from that, TMP is also found to possess anticancer, anti-oxidation, anti-radiation, and improve fatigue activities. TMP shows invaluable prospects for future applications in the form of nutraceuticals/dietary supplements. TMP has been studied worldwide, but most applications are still in the basic research stage. Although there have been many reports on the functional features of TMP, there have been relatively few studies on its specific mechanism of action. Studies on TMP properties mainly focused on its extraction and purification method, molecular weight, monosaccharide composition, side chain position, and its relationship with physiological functions. Whereas pharmacological studies of TMP are rare. Therefore, it is necessary to focus future research on the mechanism of the TMP and provide theoretical guidance for the development of TMP in food, cosmetics, medicines, and health care products. TMP has been used in some fields, it is necessary to explore the application of TMP in numerous other areas. In conclusion, TMP has great potential as a nutraceutical and functional food, as well as potentially representing a valuable source for bioactive compounds therapeutic use and pharmaceutical application.

Conflict of interest

The authors declare no conflict of interest.

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References

Agrawal, M., Bansal, S., & Chopra, K. (2021). Evaluation of the in vitro and in vivo antioxidant potentials of food grade Phycocyanin. *Journal of Food Science and Technology*, 58(11), 4382-4390. http://dx.doi.org/10.1007/s13197-020-04922-4. PMid:34538921.

Barbosa, J. R., & Carvalho, R. N. Jr. (2020). Occurrence and possible roles of polysaccharides in fungi and their influence on the development of new technologies. *Carbohydrate Polymers*, 246, 116613. http://dx.doi.org/10.1016/j.carbpol.2020.116613. PMid:32747253.

Barbosa, J. R., Freitas, M. M. S., Martins, L. H. S., & Carvalho, R. N. J. (2020). Polysaccharides of mushroom Pleurotus spp.: new extraction techniques, biological activities and development of new technologies. *Carbohydrate Polymers*, 229, 115550. http://dx.doi.org/10.1016/j.carbpol.2019.115550. PMid:31826512.

Byeon, S. E., Lee, J., Lee, E., Lee, S. Y., Hong, E. K., Kim, Y. E., & Cho, J. Y. (2009). Functional activation of macrophages, monocytes and splenic lymphocytes by polysaccharide fraction from Tricholoma matsutake. *Archives of Pharmacal Research*, 32(11), 1565-1572. http://dx.doi.org/10.1007/s12272-009-2108-y. PMid:20091269.

Carmona-Gutierrez, D., Zimmermann, A., Kainz, K., Pietrocola, F., Chen, G., Maglioni, S., Schiavi, A., Nah, J., Mertel, S., Beuschel, C. B., Castoldi, F., Sica, V., Trausinger, G., Raml, R., Sommer, C., Schroeder, S., Hofer, S. J., Bauer, M. A., Pendl, T., Tadic, J., Dammbrueck, C., Hu, Z., Ruckenstuhl, C., Eisenberg, T., Durand, S., Bossut, N., Aprahamian, F., Abdellatif, M., Sedej, S., Enot, D. P., Wolinski, H., Dengjel, J., Kepp, O., Magnes, C., Sinner, F., Pieber, T. R., Sadoshima, J., Ventura, N., Sigrist, S. J., Kroemer, G., & Madeo, F. (2019). The flavonoid 4,4′-dimethoxychalcone promotes autophagy-dependent longevity across species. *Nature Communications*, 10(1), 651. http://dx.doi.org/10.1038/s41467-019-08555-w. PMid:30783116.

Chen, Y., Du, X. J., Zhang, Y., Liu, X. H., & Wang, X. D. (2017). Ultrasound extraction optimization, structural features, and antioxidant activity of polysaccharides from Tricholoma matsutake. *Journal of Zhejiang University-Science B*, 18(8), 674-684. http://dx.doi.org/10.1631/jzus.B1600239. PMid:28786242.

Cheng, H., Jia, Y., Wang, L., Liu, X., Liu, G., Li, L., & He, C. (2016). Isolation and structural elucidation of a novel homogenous polysaccharide from Tricholoma matsutake. *Natural Product Research*, 30(1), 58-64. http://dx.doi.org/10.1080/14786419.2015.1034711. PMid:26208132.

Cheng, H., Liu, Y. H., & Liu, X. Y. (2013). Separation and purification as well as whitening efficacy of polysaccharide from Tricholoma Matsutake Sing. *China Surfactant Detergent and Cosmetics*, 43(2), 134-138.

Christopher, T., Mahamat, O., Chungong, M. N., Ngwa, C. A., & Samka, P. M. (2021). Immunological activities of crude polysaccharides extracts of Climacodon pulcherrimus (Phanerochaetaceae) in lipopolysaccharide-induced rat macrophages and neutrophils' responses. *Journal of Medicinal Food*, 24(2), 180-187. http://dx.doi.org/10.1089/jmf.2020.0005. PMid:32397875.

Cui, F. J., Qian, L. S., Sun, W. J., Zhang, J. S., Yang, Y., Li, N., Zhuang, H. N., & Wu, D. (2018). Ultrasound-assisted extraction of polysaccharides from Volvariella volvacea: process optimization and structural characterization. *Molecules*, 23(7), 1706. http://dx.doi.org/10.3390/molecules23071706. PMid:30011781.

Delazar, A., Nahar, L., Hamedeyazdan, S., & Sarker, S. D. (2012). Microwave-assisted extraction in natural products isolation. *Methods* *in Molecular Biology*, 864, 89-115. http://dx.doi.org/10.1007/978-1-61779-624-1_5. PMid:22367895.

Ding, X., Li, J., Hou, Y., & Hou, W. (2016). Comparative analysis of macrophage transcriptomes reveals a key mechanism of the immunomodulatory activity of Tricholoma matsutake polysaccharide. *Oncology Reports*, 36(1), 503-513. http://dx.doi.org/10.3892/or.2016.4814. PMid:27221808.

Ding, X., Tang, J., Cao, M., Guo, C.-X., Zhang, X., Zhong, J., Zhang, J., Sun, Q., Feng, S., Yang, Z.-R., & Zhao, J. (2010). Structure elucidation and antioxidant activity of a novel polysaccharide isolated from Tricholoma matsutake. *International Journal of Biological Macromolecules*, 47(2), 271-275. http://dx.doi.org/10.1016/j.ijbiomac.2010.04.010. PMid:20430053.

Du, B., Bian, Z., & Xu, B. (2014). Skin health promotion effects of natural beta-glucan derived from cereals and microorganisms: a review. *Phytotherapy Research*, 28(2), 159-166. http://dx.doi.org/10.1002/ptr.4963. PMid:23494974.

Du, X., Mu, H., Zhou, S., Zhang, Y., & Zhu, X. (2013). Chemical analysis and antioxidant activity of polysaccharides extracted from Inonotus obliquus sclerotia. *International Journal of Biological Macromolecules*, 62, 691-696. http://dx.doi.org/10.1016/j.ijbiomac.2013.10.016. PMid:24145301.

Ebina, T. (2003). Activation of antitumor immunity by intratumor injection of biological preparations. *Cancer & Chemotherapy*, 30(11), 1555-1558. PMid:14619462.

Geng, X., Tian, G., Zhang, W., Zhao, Y., Zhao, L., Wang, H., & Ng, T. B. (2016). A Tricholoma matsutake peptide with angiotensin converting enzyme inhibitory and antioxidative activities and antihypertensive effects in spontaneously hypertensive rats. *Scientific Reports*, 6(1), 24130. http://dx.doi.org/10.1038/srep24130. PMid:27052674.

Gong, P., Wang, S., Liu, M., Chen, F., Yang, W., Chang, X., Liu, N., Zhao, Y., Wang, J., & Chen, X. (2020). Extraction methods, chemical characterizations and biological activities of mushroom polysaccharides: a mini-review. *Carbohydrate Research*, 494, 108037. http://dx.doi.org/10.1016/j.carres.2020.108037. PMid:32592937.

Hou, Y., Ding, X., Hou, W., Song, B., & Yan, X. (2017). Structure elucidation and antitumor activity of a new polysaccharide from Maerkang Tricholoma matsutake. *International Journal of Biological Sciences*, 13(7), 935-948. http://dx.doi.org/10.7150/ijbs.18953. PMid:28808425.

Hou, Y., Ding, X., Hou, W., Zhong, J., Zhu, H., Ma, B., Xu, T., & Li, J. (2013). Anti-microorganism, anti-tumor, and immune activities of a novel polysaccharide isolated from Tricholoma matsutake. *Pharmacognosy Magazine*, 9(35), 244-249. http://dx.doi.org/10.4103/0973-1296.113278. PMid:23930009.

Jia, L. M., Liu, L., Dong, Q., & Fang, J. N. (2004). Structural investigation of a novel rhamnoglucogalactan isolated from the fruiting bodies of the fungus Hericium erinaceus. *Carbohydrate Research*, 339(16), 2667-2671. http://dx.doi.org/10.1016/j.carres.2004.07.027. PMid:15519325.

Jing, H., Li, J., Zhang, J., Wang, W., Li, S., Ren, Z., Gao, Z., Song, X., Wang, X., & Jia, L. (2018). The antioxidative and anti-aging effects of acidic- and alkalic-extractable mycelium polysaccharides by Agrocybe aegerita (Brig.) Sing. *International Journal of Biological Macromolecules*, 106, 1270-1278. http://dx.doi.org/10.1016/j.ijbiomac.2017.08.138. PMid:28851637.

Leong, Y. K., Yang, F. C., & Chang, J. S. (2021). Extraction of polysaccharides from edible mushrooms: emerging technologies and recent advances. *Carbohydrate Polymers*, 251, 117006. http://dx.doi.org/10.1016/j.carbpol.2020.117006. PMid:33142573.

Li, M., Dong, L., Du, H., Bao, Z., & Lin, S. (2021). Potential mechanisms underlying the protective effects of Tricholoma matsutake singer peptides against LPS-induced inflammation in RAW264.7 macrophages. *Food Chemistry*, 353, 129452. http://dx.doi.org/10.1016/j.foodchem.2021.129452. PMid:33714115.

Li, Q. Z., Wu, D., Zhou, S., Liu, Y. F., Li, Z. P., Feng, J., & Yang, Y. (2016). Structure elucidation of a bioactive polysaccharide from fruiting bodies of Hericium erinaceus in different maturation stages. *Carbohydrate Polymers*, 144, 196-204. http://dx.doi.org/10.1016/j.carbpol.2016.02.051. PMid:27083809.

Li, Q., Wang, Y., Cai, G., Kong, F., Wang, X., Liu, Y., Yang, C., Wang, D., & Teng, L. (2015). Antifatigue activity of liquid cultured Tricholoma matsutake mycelium partially via regulation of antioxidant pathway in mouse. *BioMed Research International*, 2015, 562345. http://dx.doi.org/10.1155/2015/562345. PMid:26697489.

Li, Q., Wang, Y., Chen, Y., Teng, M., He, J., Wang, X., Kong, F., Teng, L., & Wang, D. (2017). Investigation of the immunomodulatory activity of Tricholoma matsutake mycelium in cyclophosphamide-induced immunosuppressed mice. *Molecular Medicine Reports*, 16(4), 4320-4326. http://dx.doi.org/10.3892/mmr.2017.7090. PMid:28765877.

Li, X., He, Y., Zeng, P., Liu, Y., Zhang, M., Hao, C., Wang, H., Lv, Z., & Zhang, L. (2019). Molecular basis for Poria cocos mushroom polysaccharide used as an antitumour drug in China. *Journal of Cellular and Molecular Medicine*, 23(1), 4-20. http://dx.doi.org/10.1111/jcmm.13564. PMid:30444050.

Liu, G., Wang, H., Zhang, H., & Zhou, B. H.(2013). Study of antitumor activities of polysaccharides from Tricholoma Matsutake in vitro. *Chinese Archives of Traditional Chinese Medicine*, 31(2), 267-270.

Marić, M., Grassino, A. N., Zhu, Z., Barba, F. J., Brnčić, M., & Brnčić, S. R. (2018). An overview of the traditional and innovative approaches for pectin extraction from plant food wastes and by-products: ultrasound-, microwaves-, and enzyme-assisted extraction. *Trends in Food Science & Technology*, 76, 28-37. http://dx.doi.org/10.1016/j.tifs.2018.03.022.

Marzuki, S. U., Pranoto, Y., Khumsap, T., & Nguyen, L. T. (2021). Effect of blanching pretreatment and microwave-vacuum drying on drying kinetics and physicochemical properties of purple-fleshed sweet potato. *Journal of Food Science and Technology*, 58(8), 2884-2895. http://dx.doi.org/10.1007/s13197-020-04789-5. PMid:34294950.

Ren, L., Zhang, J., & Zhang, T. (2021). Immunomodulatory activities of polysaccharides from Ganoderma on immune effector cells. *Food Chemistry*, 340, 127933. http://dx.doi.org/10.1016/j.foodchem.2020.127933. PMid:32882476.

Ren, Y., Bai, Y., Zhang, Z., Cai, W., & Flores, A. R. (2019). The preparation and structure analysis methods of natural polysaccharides of plants and fungi: a review of recent development. *Molecules*, 24(17), 3122. http://dx.doi.org/10.3390/molecules24173122. PMid:31466265.

Tong, H., Liu, X., Tian, D., & Sun, X. (2013). Purification, chemical characterization and radical scavenging activities of alkali-extracted polysaccharide fractions isolated from the fruit bodies of Tricholoma matsutake. *World Journal of Microbiology & Biotechnology*, 29(5), 775-780. http://dx.doi.org/10.1007/s11274-012-1232-x. PMid:23242547.

Veeraperumal, S., Qiu, H. M., Tan, C. S., Ng, S. T., Zhang, W., Tang, S., Cheong, K. L., & Liu, Y. (2021). Restitution of epithelial cells during intestinal mucosal wound healing: the effect of a polysaccharide from the sclerotium of Lignosus rhinocerotis (Cooke) Ryvarden. *Journal of Ethnopharmacology*, 274, 114024. http://dx.doi.org/10.1016/j.jep.2021.114024. PMid:33727110.

Wang, H. (2008). Protective effect of Tricholoma Matsutake polysaccharide on immune function of radiation-injured mice. *Journal of Xi'an Jiaotong University*, 31(3), 388-390.

Wang, J., Cao, B., Zhao, H., & Feng, J. (2017). Emerging roles of Ganoderma Lucidum in anti-aging. *Aging and Disease*, 8(6), 691-707. http://dx.doi.org/10.14336/AD.2017.0410. PMid:29344411.

Wang, N., Dai, L., Chen, Z., Li, T., Wu, J., Wu, H., Wu, H., & Xiang, W. (2021a). Extraction optimization, physicochemical characterization, and antioxidant activity of polysaccharides from Rhodosorus sp. SCSIO-45730. *Journal of Applied Phycology*, 1-15. Ahead of print. PMid:34866795.

Wang, Q. (2015). Study on the whitening activity and pathway of Tricholoma matsutake TMSP-5II. *Natural Product Research and Development*, 27(1), 134-138.

Wang, Q., Zhao, Y., Feng, X., Ibrahim, S. A., Huang, W., & Liu, Y. (2021b). Effects of drying on the structural characteristics and antioxidant activities of polysaccharides from Stropharia rugosoannulata. *Journal of Food Science and Technology*, 58(9), 3622-3631. http://dx.doi.org/10.1007/s13197-021-05120-6. PMid:34366479.

Wang, W., Xue, C., & Mao, X. (2020). Radioprotective effects and mechanisms of animal, plant and microbial polysaccharides. *International Journal of Biological Macromolecules*, 153, 373-384. http://dx.doi.org/10.1016/j.ijbiomac.2020.02.203. PMid:32087223.

Wang, X.-Y., Zhang, D.-D., Yin, J.-Y., Nie, S.-P., & Xie, M.-Y. (2019). Recent developments in Hericium erinaceus polysaccharides: extraction, purification, structural characteristics and biological activities. *Critical Reviews in Food Science and Nutrition*, 59(Suppl. 1), S96-S115. http://dx.doi.org/10.1080/10408398.2018.1521370 . PMid:30421988.

Wu, D. T., Li, W. Z., Chen, J., Zhong, Q. X., Ju, Y. J., Zhao, J., Bzhelyansky, A., & Li, S. P. (2015). An evaluation system for characterization of polysaccharides from the fruiting body of Hericium erinaceus and identification of its commercial product. *Carbohydrate Polymers*, 124, 201-207. http://dx.doi.org/10.1016/j.carbpol.2015.02.028. PMid:25839812.

Xu, L., Zhu, M., Chen, X., Wang, H., & Zhang, G. (2015). A novel laccase from fresh fruiting bodies of the wild medicinal mushroom Tricholoma matsutake. *Acta Biochimica Polonica*, 62(1), 35-40. http://dx.doi.org/10.18388/abp.2014_713. PMid:25781157.

Yao, H. W., Zhang, X. Y., & Feng, L. (2008). Microwave and ultrasound extraction technology polysaccharides of Tricholoma Matsutake. *Forest By-Product and Speciality in China*, 2008(6), 10-15.

Yin, M., Zhang, Y., & Li, H. (2019). Advances in research on immunoregulation of macrophages by plant polysaccharides. *Frontiers in Immunology*, 10, 145. http://dx.doi.org/10.3389/fimmu.2019.00145. PMid:30804942.

Yin, X. L., You, Q. H., & Jiang, Z. H. (2011). Different solvent extracts of Tricholoma matsutake and their antioxidant activity. *Shipin Yu Fajiao Gongye*, 37(5), 194-198.

Yin, X., You, Q., & Jiang, Z. (2012). Immunomodulatory activities of different solvent extracts from Tricholoma matsutake (S. Ito et S. Imai) singer (higher basidiomycetes) on normal mice. *International Journal of Medicinal Mushrooms*, 14(6), 549-556. http://dx.doi.org/10.1615/IntJMedMushr.v14.i6.20. PMid:23510248.

Yin, X., You, Q., & Su, X. (2014). A comparison study on extraction of polysaccharides from Tricholoma matsutake by response surface methodology. *Carbohydrate Polymers*, 102, 419-422. http://dx.doi.org/10.1016/j.carbpol.2013.11.072. PMid:24507300.

You, L., Gao, Q., Feng, M., Yang, B., Ren, J., Gu, L., Cui, C., & Zhao, M. (2013). Structural characterisation of polysaccharides from Tricholoma matsutake and their antioxidant and antitumour activities. *Food Chemistry*, 138(4), 2242-2249. http://dx.doi.org/10.1016/j.foodchem.2012.11.140. PMid:23497882.

Yuan, S., Xu, C. Y., Xia, J., Feng, Y. N., Zhang, X. F., & Yan, Y. Y. (2020). Extraction of polysaccharides from Codonopsis pilosula by fermentation with response surface methodology. *Food Science & Nutrition*, 8(12), 6660-6669. http://dx.doi.org/10.1002/fsn3.1958. PMid:33312549.

Zeng, W. C., Zhang, Z., Gao, H., Jia, L. R., & Chen, W. Y. (2012). Characterization of antioxidant polysaccharides from Auricularia auricular using microwave-assisted extraction. *Carbohydrate Polymers*, 89(2), 694-700. http://dx.doi.org/10.1016/j.carbpol.2012.03.078. PMid:24750775.

Zhao, Y. M., Yang, J. M., Liu, Y. H., Zhao, M., & Wang, J. (2018). Ultrasound assisted extraction of polysaccharides from Lentinus edodes and its anti-hepatitis B activity in vitro. *International Journal of Biological Macromolecules*, 107(Pt B), 2217-2223. http://dx.doi.org/10.1016/j.ijbiomac.2017.10.100. PMid:29051096.

Zhao, Z., Xu, X., Ye, Q., & Dong, L. (2013). Ultrasound extraction optimization of Acanthopanax senticosus polysaccharides and its antioxidant activity. *International Journal of Biological Macromolecules*, 59, 290-294. http://dx.doi.org/10.1016/j.ijbiomac.2013.04.067. PMid:23628583.

Zheng, S., Zhang, G., Wang, H., Long, Z., Wei, T., & Li, Q. (2021). Progress in ultrasound-assisted extraction of the value-added products from microorganisms. *World Journal of Microbiology & Biotechnology*, 37(4), 71. http://dx.doi.org/10.1007/s11274-021-03037-y. PMid:33763773.

Zhu, H., Yuan, Y., Liu, J., Zheng, L., Chen, L., & Ma, A. (2016a). Comparing the sugar profiles and primary structures of alkali-extracted water-soluble polysaccharides in cell wall between the yeast and mycelial phases from Tremella fuciformis. *Journal of Microbiology*, 54(5), 381-386. http://dx.doi.org/10.1007/s12275-016-5533-x. PMid:27095457.

Zhu, Y., Yu, X., Ge, Q., Li, J., Wang, D., Wei, Y., & Ouyang, Z. (2020). Antioxidant and anti-aging activities of polysaccharides from Cordyceps cicadae. *International Journal of Biological Macromolecules*, 157, 394-400. http://dx.doi.org/10.1016/j.ijbiomac.2020.04.163. PMid:32339570.

Zhu, Z.-Y., Dong, F., Liu, X., Lv, Q., YingYang, Liu, F., Chen, L., Wang, T., Wang, Z., & Zhang, Y. (2016b). Effects of extraction methods on the yield, chemical structure and anti-tumor activity of polysaccharides from Cordyceps gunnii mycelia. *Carbohydrate Polymers*, 140, 461-471. http://dx.doi.org/10.1016/j.carbpol.2015.12.053. PMid:26876874.