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Critical Review

Beta-glucans from edible and medicinal mushrooms: Characteristics, physicochemical and biological activities

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ABSTRACT

One of the main active components from mushrooms was recently identified as β-glucan. Health-promoting β-glucans are an auspicious group of polysaccharides. β-Glucans from different sources such as cereals, yeast and grass have previously been documented. However, information on mushroom β-glucan is limited. This review summarizes the extraction, purification, quantification, and structural characterization of β-glucans, along with chemical and biological activities from this compound from mushrooms, and the current status of this research area with a view for future directions.

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1. Introduction

Mushroom is the spore-bearing fruiting body of a fungus, typically produced above ground on soil. Fresh and preserved mushrooms are consumed in many countries as a delicacy, particularly for their specific aroma and texture (Kalac, 2013). It is well-known that mushrooms contain a very large variety of biomolecules with nutritional (Kalac, 2009) and medicinal properties (Borchers et al., 2004; Lindequist et al., 2005; Poucheret et al., 2006). Due to these properties, mushrooms have been recognized as natural sources for the development of medicines and nutraceuticals (Alves et al., 2012).

Mushrooms are also recognized as functional foods for their bioactive compounds that offer diverse beneficial impacts on human health. One such potent component is β-glucan (Ren et al., 2012). Most β-glucans are derived from the fruiting bodies of mushrooms. β-Glucans are polysaccharides of D-glucose monomers linked by β-glycosidic bonds. It is a kind of dietary fibre in cereals, yeasts, mushrooms, some bacteria and seaweeds (Du et al., 2013). β-Glucan from mushrooms consists of β-(1 → 3) and (1 → 6) linkages (Fig. 1) (Laroche and Michaud, 2007). β-Glucans from various sources have different branching pattern, linkage type and molecular weight (MW). β-Glucans from mushroom comprise a backbone of glucose residues linked by β-(1 → 3)-glycosidic bonds with attached β-(1 → 6) branch points, which exhibits antitumour and immune-stimulating properties. Mushroom-originated β-glucan seems to show very different activities from β-glucan derived from oat and barley. Mushroom β-glucan has shown effectiveness as an anti-tumour defense and as an immune system booster. β-Glucan from oats and barley help in lowering cholesterol and blood sugar. The primary structure, solubility, degree of branching (DB), MW, charge of polymers, and structure in aqueous media, all affect biological activities of β-glucan (Zekovic et al., 2005).

Review papers about β-glucan from different sources, such as cereals, yeast, and grass, have been published (Buckeridge et al., 2004; Brennan and Cleary, 2005; Lazaridou and Biliaderis, 2007; Wood, 2007; Petravic-Tominac et al., 2010; Havrlentová et al., 2011; Daou and Zhang, 2012; Rieder and Samuelsen, 2012). However, the review of mushroom β-glucan has received little attention. Thus, the main objective of this review is to address recent research findings on sources, structural features, viscosity and molecular weight, extraction, purification, and detection methods of β-glucan from edible and medicinal mushrooms. The biological properties of β-glucan from edible and medicinal mushrooms have also been included in this contribution.

2. Occurrence of β-glucan in mushroom

Mushrooms major cultivated species also contain relatively large amounts of carbohydrate and fibre, ranging from 51% to 88% and from 4% to 20% (dry weight basis) (Mattila et al., 2000). Shiitake (*Lentinus edodes*) and some members of the genus Oyster (*Pleurotus* spp.) are acknowledged to be some of the most important sources of β-glucans (Rop et al., 2009). Lentinan from the fruiting body of shiitake is a representative mushroom (1 → 3)-β-glucan with effective antitumour and immunopotentiating activity. Its primary structure is a (1 → 3)-β-glucan consisting of five (1 → 3)-β-glucose residues in a linear linkage and two (1 → 6)-β-glucopyranoside branches in side chains which result in a right-handed triple helical structure (Chihara, 2001). Another highly potent antitumour polysaccharide, schizophyllan from mushroom *Schizophyllum commune* is also a (1 → 3)-β-glucan having a β-glucopyranosyl group linked 1 → 6 to every third or fourth residue of the main chain. It is similar to lentinan in its triple helix structure and biological activities, but physico-chemically differs from lentinan. β-Glucans are also present in many other mushrooms, such as *Ganoderma lucidum*, *Grifola frondosa*, *Pleurotus abalones*, *Flammulina velutiper* and *Auricularia auricula*. A survey of the most important biologically active glucan and of the mushrooms in which they occur is presented in Table 1.

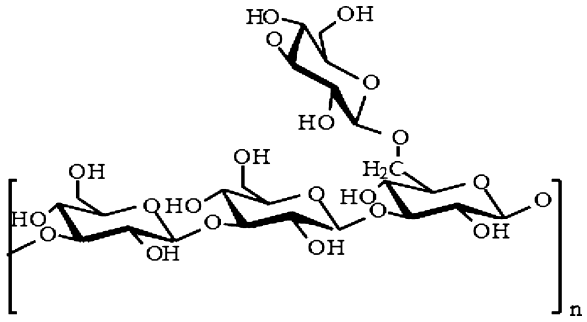

Fig. 1. Structure of (1 → 3) β-glucans with ramifications β (1 → 6). Copyright 2007 Recent Patents on Biotechnology. (Laroche and Michaud, 2007. New developments and prospective applications for β-(1,3) glucans. (Recent Patents on Biotechnology 1: 59–73).

3. Extraction and production of mushroom β-glucan

Extraction of β-glucan is a difficult job and requires special attention to produce consistent raw material (Ahmad et al., 2012). The nature of extraction procedure has a profound effect on the MW of β-glucan, which in turn affects its functional behaviour (Brennan and Cleary, 2005). The extraction methodologies are based on the solubility of β-glucan in hot water and in alkaline solutions. Separation of the dissolved proteins by isoelectric precipitation, and precipitation of the β-glucan by ammonium sulphate, 2-propanol, or ethanol are general ways. Kim et al. (2005) used distilled water (100 °C) at a ratio of 1:10 (w/v) to extract β-glucan from *Agaricus blazei* with extraction time 3 h. They reported that β-glucan showed anti-hyperglycaemic, anti-hypertriglyceridemic, anti-hypercholesterolemic, and anti-arteriosclerotic activities, thus indicating anti-diabetic activity as a whole in diabetic rats. Kumari et al. (2008) determined optimal operating conditions using one factor at a time method and response surface method to maximize production of schizophyllan by *S. commune* from an initial value of 1.06 g L^{-1} to 8.06 g L^{-1}. The fermentation was carried out for 168 h at 28 ± 2 °C on an orbital shaker at 180 rpm. Moreover, Kao et al. (2012) had successfully isolated low-molecular-weight β-1,3-glucan, in high yields, from the waste residue of extracted fruiting bodies of *G. lucidum*. They examined the polysaccharide from the residues of alkaline-extracted fruiting bodies using high-performance anion-exchange chromatography. The extraction techniques of polysaccharides (including β-glucan) have been summarized in a latest report of Villares et al. (2012).

4. Characteristics of mushroom β-glucans

4.1. Structural features of mushroom β-glucan

β-Glucans are cell wall components of mycete. Structural diversity of fungal (including mushrooms) glucans has been reviewed in a recent report (Synytsya and Novak, 2013). Structurally, mushroom β-glucans consist of linear β-(1 → 3)-linked backbones with β-(1 → 6)-linked side chains of varying

Table 1
Extraction, purification and structural features of β-glucans from different mushroom sources.

Source	Name of β-glucans	Extraction process	Purification	Structural features	References
Himematsutake mushroom (*Agaricus brasiliensis*)	β-Glucan	Water extraction (100 °C)	DEAE-cellulose column chromatography; Toyopear HW-65F column; Con A-Sepharose 4B column	A greater proportion of (1 → 6)-β-side branches on the (1 → 3)-β-backbone	Camelini et al. (2005)
Common pelit gill (*Schizophyllum commune*)	Schizophylan	Hot water extraction (HWE); Hot alkali extraction (HAE)	Thin-layer chromatography	A mixture of α/β glucose in the ratio 1/1.3; Amixture of α/β glucose at a 1/1 ratio, and α/β mannose at 0.6/0.4 ratio *vs* α-glucose	Klaus et al. (2011)
Reishi (*Ganodema lucidum*)	Ganoderan	Water extraction (100 °C) and graded ethanol precipitation	DEAE-cellulose column and Sephacryl S-300 column	A 1,6-linked β-D-glucopyranosyl backbone with different length of branches consisting of terminal and 1,4-linked glucopyranosyl residues, attached to *O*-4 of alternative glucose residues in the backbone	Dong et al. (2012)
Shiitake (*Lentinus edodes*)	Lentinan	Water extraction and absolute ethanol precipitation	–	Highly branched glucan containing mainly 1, 3 and 1, 6 linkages	Rasmy et al. (2010)
		Dynamic high pressure microfluidization extraction	–	–	Huang et al. (2012)
Oyster mushroom (*Pleurotus ostreatus*)	Pleuran	80% Methanol extraction; Neutral saline (0.9% NaCl) extraction; Water extraction (100 °C)	–	–	Cha et al. (2012)
Maitake (*Grifola fondosa*)	Grifolan	Water extraction (100 °C), ethanol precipitation, alkaline extraction, acid precipitation and protein depletion	–	–	Masuda et al. (2012)
		Alkali (5% NaOH) extraction	DEAE-cellulose column	A β-D-(1-3)-linked glucan backbone with a single β-D-(1-6)-linked glucopyranosyl residue branched at C-6 on every third residue.	Fang et al. (2012)
Rainbow conk (*Coriolus versicolor*)	Krestin	Hot water extraction	DEAE-cellulose anion-exchange chromatography with 0.7 M NaCl	-	Kang et al. (2013)
Enokitake mushroom (*Flammulina velutipes*)	Flammulin	Aqueous and alkaline extraction	–	A main chain of (1 → 3)-linked-glucopyranosyl residues, substituted at *O*-6 by single-unit β- glucopyranosyl side chains	Smiderle et al. (2006)
Judas's ear (*Auricularia auricula*)	β-Glucan	70% Ethanol/water solution extraction	–	A main chain of (1 → 4)-linked D-glucopyranosyl with glucopyranosyl side groups at *O*-6	Ma et al. (2008)
		0.15 M Aqueous NaCl extraction (80–100 °C)	–	A β-(1 → 3)-D-glucan with two β-(1 → 6)-D-glucosyl residues for every three main chain glucose residues	Xu et al. (2012)
Honey fungus (*Armillaria mellea*)	β-Glucan	Water extraction	–	–	Lai and Ng (2013)
Bamboo fungus (*Dictyophora indusiata*)	β-Glucan	Water extraction (90 °C)	Sephadex G-200 gel chromatography	A backbone of β-conformation, mainly consist of glucose (98.58%)	Deng et al. (2012)
Geastrum saccatum mushroom (*Geastrum saccatum*)	β-Glucan	Water extraction (100 °C)	–	High amount of glucose and traces of galactose. The signal appearing at 103.5 ppm was assigned to C1 of β-glucose.	Guerra Dore et al. (2007)
Tuckahoe (*Poria cocos*)	β-Glucan	Water extraction	–	(1 → 3)-β-D-glucan with some β-(1 → 6) linked branches; (1 → 3)-β-D-glucan containing some glucose branches	Wang et al. (2004)
		Hot water, 0.5 M NaOH and 88% formic acid extraction	–	(1 → 3)-β-D-glucans	Jin et al. (2003)
Pleurotus sajor-caju	β-Glucan	Hot aqueous extraction	–	A branched structure with a (1 → 3)-linked β-Glcp main-chain, substituted at O-6 by single-unit β-Glcp side-chains	Carbonero et al. (2012)
		Hot water extraction	DEAE Toyopearl and Sepharose CL-6B column chromatography	β-(1 → 3)(1 → 6)-glucan in the native triple helical structure	Satitmanwiwat et al. (2012)
Hybrid mushroom of *Pleurotus florida* and *Volvariella volvacea*	β-Glucan	Water extraction	Sepharose 6B column	A (1 → 6)-β-D-glucan →6)-β-D-Glc*p*-(1→6)-β-D-Glc*p*-(1→6)-β-D-Glc*p*-(1→ 3 ↑ 1 β-D-Glc*p*	Das et al. (2010)
		Alkaline extraction	DEAE-cellulose column and Sepharose 6B column		Maity et al. (2013)

length and distribution, and can form complex tertiary structures stabilized by interchain hydrogen bonds (Brown and Gordon, 2005). Both schizophyllan from mushroom *S. commune* and scleroglucan from mushroom *Sclerotium glucanicum* have a β-(1 → 3)-linked backbone, with on average one β-(1 → 6)-glucose substitution every three backbone residues. Epiglucan from mushroom *Epicoccum nigrum* also has a backbone of β-(1 → 3)-linked glucose residues, but now with two β-(1 → 6) substitutions on average every three residues, while the β-(1 → 3)-linked backbone of lentinan from mushroom *L. edodes* has two β-(1 → 6) side chains every five residues (Schmid et al., 2001). In addition, five water-soluble heteropolysaccharides designated GL-I and GL-V, were isolated from the cultured fruit body of *G. lucidum*. As successive isolations proceed to give GL-I to GL-V, the content of glucan increased, the content of galactan and mannan decreased, and the branching decreased from high to near linear. GL-V is mainly a linear glucan (Wang et al., 2011).

In the study of Fang et al. (2012), the structural characterization of β-glucan from mushroom *G. frondosa* was emphasized by infrared spectroscopy, nuclear magnetic resonance (NMR), methylation and monosaccharide composition analysis. Its structure was determined to be a β-D-(1-3)-linked glucan backbone with a single β-D-(1-6)-linked glucopyranosyl residue branched at C-6 on every two residues. Dong et al. (2012) characterized the chemical structure of β-glucan (GLSA50-1B) from mushroom *G. lucidum* using sugar compositional analysis, methylation analysis, partial acid hydrolysis, acetolysis, and NMR and ESI-MS spectroscopy. The results indicated that GLSA50-1B was elucidated to be a novel β-D-glucan featured by a 1,6-linked β-D-glucopyranosyl backbone with different length of branches consisting of terminal and 1,4-linked glucopyranosyl residues, attached to *O*-4 of alternative glucose residues in the backbone. Sen et al. (2013) emphasized that the structural characterization of β-glucan from a hybrid mushroom, *pfls1h* of *Pleurotus florida* and *Lentinus squarrosulus* (Mont.) Singer, using total hydrolysis, methylation analysis, peroxide oxidation, and NMR techniques (^{1}H, ^{13}C, DEPT-135, DQF-COSY, TOCSY, NOESY, ROESY, HSQC, and HMBC). Methylation analysis revealed that purified polysaccharide fraction (PS-I) was composed of (1 → 3, 6), (1 → 3), (1 → 6)-linked and terminal β-D-glucopyranosyl residues in a relative proportion of approximately 1:1:1:1. The repeating unit of the glucan consists of a backbone chain of two (1 → 6)-β-D-glucopyranosyl residues, one of which is branched at *O*-3 position with (1 → 3)-β-D-glucopyranosyl and terminated with a β-D-glucopyranosyl residue.

The content of β-glucan can be influenced by many factors, such as species, growing conditions of mushrooms, the degree of fruiting body maturity and total dietary fibre content and so on. Kyanko et al. (2013) evaluated that total dietary fibre of isolates exhibited a noticeable variability from 16% to 53% and the highest values were obtained from the genera *Paecilomyces* and *Penicillium*, a fact consistent with a higher content of β-glucans (24% and 17%, respectively), higher than previously reported for Basidiomycetes and yeast. The structural features of β-glucan from their respective sources are shown in Table 1.

4.2. Solubility, viscosity and molecular weight of mushroom β-glucan

The solubility of β-glucans in water is dependent, above all, on their structure, and this is associated with their origin as well (Rop et al., 2009). It is well known that the solubility of β-glucan increases with temperature. In general, water solubility chain conformation, and introduction of suitable ionic groups with appropriate degree of substitution can change the bioactivities of polysaccharides (Tao et al., 2006). Water-insoluble polysaccharides show little bioactivity, whereas their water-soluble sulfated derivatives exhibit high antitumor and/or antiviral activities (Zhang et al., 2001). Water solubility and introduction of sulphate groups were the main factors in enhancing the antitumor activities. Tao et al. (2006) reported the sulphated derivatives exhibited relatively higher *in vitro* antitumour activity against human hepatic cancer cell line HepG2 than the native water-soluble hyper-branched β-glucan. Soluble β-glucans appeared to be stronger immuno-stimulators than insoluble β-glucans. The reasons for this are not totally clear. Moreover, the exact mechanism of intestinal absorption of orally administrated β-glucan remains unclear.

The viscosity of β-glucan is dependent on the molecular weight, molecular structure, and food matrix (Kerckhoffs et al., 2003; Bae et al., 2009). The range of relative MW of β-glucans is quite wide and fluctuates (depending on origin) from tens to thousands of kilodaltons (kDa) (Rop et al., 2009).

Oat and mushroom β-glucans in the diet have been reported to lower human plasma lipid, one of the mechanisms of oat β-glucan in lowering plasma lipid is explained to be its capacity to increase food viscosity (Bae et al., 2009; Andersson et al., 2010). Researchers found that β-glucans from both oat and mushroom shiitake exhibited sheer thinning behaviour where the viscosity decreased with higher speed. They also found that the mushroom diet was better than the oat diet as part of a high fat diet because it reduced body weight gain, total fat mass, plasma triacylglycerol and increased fat foecal excretion (Handayani et al., 2012). Moreover, viscosity is controlled by β-glucan concentration in solution and β-glucan MW, and therefore the glycaemic response was significantly correlated to values of concentration × MW (Wood et al., 2000). Wood (2007) presented a review focused on the relationship of viscosity and MW of cereal β-glucan in postproandial blood glucose and insulin response and serum cholesterol.

Size-exclusion chromatography (SEC) is a chromatographic method in which molecules in solution are separated by molecular size or molecular weight (Stephanie et al., 2007). SEC is a widely used characterization method for polysaccharides because of its ability to provide good molar mass distribution results for polysaccharides. This technique was used in combination with static laser light scattering and low-angle laser-light-scattering for determining MW of β-glucan from different mushrooms, such as mushroom *Cryptoporus volvatus*, mushroom *Poria cocos* (Wang et al., 2004), mushroom *A. auricula* (Ma et al., 2008; Xu et al., 2012). When an aqueous solution is used to elute the sample through the column, SEC is known as gel filtration chromatography (GFC), as opposed to the name gel permeation chromatography (GPC), which is used when an organic solvent is used as a mobile phase. In the study of Dong et al. (2012), a homogeneous polysaccharide from mushroom *Ganodema lucidum* was characterized as a β-D-glucan with a molecular weight of 103 kDa using high performance gel permeation chromatography (HPGPC). Moreover, Fang et al. (2012) obtained a soluble homogeneous β-glucan with a molecular mass of 300 kDa using HPGPC from mushroom *Grifola fondosa*. Kang et al. (2013) reported the molecular weight of β-glucan from another mushroom *Coriolus versicolor* after Sepharose CL-4B GFC was about 750 kDa. Table 2 presents a brief review of different MW and techniques used for MW determination of β-glucan from their respective sources. The role of MW in the pharmaceutical activity of β-glucans is very important.

5. Purification of mushroom β-glucan

Ion-exchange chromatography and gel filtration chromatograph are the most common and convenient methods for purifying polysaccharide. In general, the crude polysaccharide extracts were further applied to a Sephadex column and eluted with water. The

Table 2
Molecular weight and determination techniques of β-glucans from different mushroom sources.*.

Product name	Sources	Molecular weight (kDa)	Determination techniques	Column	Mobile phase	References
Lentinan	*Lentinus edodes*	913 and 965	GPC	TSK G-5000 PW XL	0.02 M phosphate buffer (KH_2PO_4, pH 6.0)	Huang et al. (2012)
β-Glucan	*Pleurotus sajor-caju*	975	HPSEC	Sepharose CL-4B	0.1 M $NaNO_3$ containing 0.5 g L^{-1} NaN_3	Carbonero et al. (2012)
		45	HPSEC	Sepharose CL-6B (1.5 × 60.0 cm)	Distilled water	Satitmanwiwat et al. (2012)
Ganoderan	*Ganodema lucidum*	103	HPGPC	Ultrahydrogel 500	0.003 M NaOAc	Dong et al. (2012)
Grifolan	*Grifola fondosa*	300	HPGPC	Ultrahydrogel 500	10 mM NaOH	Fang et al. (2012)
Krestin	*Coriolus versicolor*	750	GFC	Sepharose CL-4B	0.1 M $NaNO_3$ containing 0.5 g L^{-1} NaN_3	Kang et al. (2013)
β-Glucan	*Auricularia auricula*	34–288	SEC combined static laser light scattering	TSK-GEL G4000 PW XL (7.8 mm × 300 mm)	0.1 M NaCl	Ma et al. (2008)
		2070–2150	SEC combined static laser light scattering	ShodexOHpak SB-806 M HQ (8.0 mm × 300 mm)	0.9% aqueous NaCl	Xu et al. (2012)
β-Glucan	*Poria cocos*	91 211	SEC combined with laser light scattering	TSK-GEL G5000 (7.8 mm × 300 mm)	Phosphate buffer solution	Wang et al. (2004)
		1211	SEC combined with laser light scattering	TSK-GEL G5000 (7.8 mm × 300 mm)	0.5 M NaCl	Jin et al. (2003)

*GPC, gel permeation chromatography; HPSEC, high performance size exclusion chromatography; HPGPC, high performance gel permeation chromatography; GFC, gel filtration chromatography; SEC, size exclusion chromatography.

polysaccharide fraction was collected, applied to a Sephadex A-25 column, and eluted first with 20 mM Tris/HCl buffer (pH 8.0), then with a 0–0.2 M gradient of NaCl. The average MW of β-glucans was determined to be 30–50 kDa by GFC. Camellini et al. (2005) extracted β-glucan from mushroom *Agaricus brasiliensis* with boiling water, and then the crude polysaccharide was passed through a DEAE-cellulose column chromatography for further purification. However, the major drawbacks of ion exchange chromatography is its buffer requirement. This restriction may require a buffer exchange step prior to ion exchange chromatography. In another study, the dried mushroom *Flammulina velutipes* was milled and submitted to aqueous and alkaline extractions, and then the crude β-glucan residue was subjected to further alkaline extraction at 100 °C, using 2% and then 25% KOH (Smiderle et al., 2006). β-Glucan from mushroom *Sparassis crispa* was prepared using cold alkali (10% NaOH/5% urea) at 4 °C with a 2-day extraction time, and subsequently the extract was applied to a DEAE Sephadex A25 column for further purification (Shibata et al., 2012). Sen et al. (2013) also used distilled water (100 °C) to extract β-glucan from a hybrid mushroom, *pfls1h* of *P. florida* and *L. squarrosulus* (Mont.) Singer, with extraction time 8 h. The crude polysaccharide was passed through Sepharose 6B GPC using water as the eluant with a flow rate of 0.5 mL min^{-1}. Two fractions of purified polysaccharide named PS-I and PS-II were obtained. The MW of PS-I was estimated as about 194 kDa from a calibration curve of standard dextran. Their results suggest that the method is simple and workable for purification of polysaccharides. The advantages of gel filtration chromatograph include good separation of large molecules from the small molecules with a minimal volume of eluate. In summary, using a combination of ion-exchange chromatography and gel filtration chromatograph, purification of β-glucan is achieved. The extraction and purification techniques used for extraction of β-glucan from their respective sources are summarized in Table 1.

6. Quantification of mushroom β-glucan

With the advantages of modern structure elucidation, β-glucans were early identified as one of the most potent components with biological activities, such as biological response modifier activity and anti-carcinogenic activity in mushroom (Nitschke et al., 2011). Development of mushroom with greater β-glucan content would increase the nutritional and economic values of mushrooms. A reliable and rapid quantitative analysis of β-glucan is essential. However, quantification of β-glucans is still quite difficult to standardize because of large diversity of β-glucans. Therefore, we summarize the detection methods of β-glucan from mushroom as: (1) enzymic method or McCleary method (Megazyme kit), (2) enzyme-linked immunosorbent assay (ELISA) method, (3) fluorimetric method with aniline blue, and (4) colorimetric method with Congo red.

McCleary method is based on an enzymatic hydrolysis and quantification of the free sugars released (McCleary and Codd, 1991). However, this method has various practical limitations. A modified enzymic method is now the recommended AACC (2011) Method 32-23.01 and AOAC (1995) Official Method 995.16 for measurement of α-glucan in oat fractions and unsweetened oat cereals. According to Manzi and Pizzoferrato (2000), the amount of β-glucans in different species of edible mushrooms has been evaluated using the McCleary method with modification. The β-glucan contents of three different fractions extracted from the fruiting bodies of *Pleurotus nebrodensis* were quantitatively determined using a β-glucan assay kit (Cha et al., 2012). Moreover, the content of β-glucans in mushroom *C. versicolor* was determined enzymatically using a Megazyme kit (Kang et al., 2013). However, the use of enzymes is expensive and time-consuming and hydrolysis by acids is very unspecific.

An ELISA method has been developed by Mizuno et al. (2001) to determine high MW β-glucans, with grifolan and lentinan as prototypes of branched β-1,3–1,6-glucan with a triple helix for immune reactions to form specific antibodies.

Ko and Lin (2004) developed a fluorescence microassay based on aniline blue dye to measure β-glucan content in foods, including cereals, tubers, vegetables, fruits, and mushrooms. This microassay displayed selectivities among various 1,3-β-glucan species. The results indicated that biologically active ones such as pachyman from mushroom *P. cocos* and yeast glucan possessed much stronger fluorescent signals than others such as barley glucan and laminarin from *Laminaria japonica*. The main disadvantages of this method lie in its complexity and the shortbench-life of the solution used.

Nitschke et al. (2011) described a colorimetric method for β-1,3–1,6-glucan quantification based on the dye Congo red and determined the β-1,3–1,6-glucan content in mycelia and fruiting

bodies from various mushrooms, such as button mushroom, enokitake, maitake, shiitake, and shimeji mushroom, among others. Congo red was used for characterization of glucan tertiary structures because of its interactions with the triple helix of β-1,3–1,6-glucan (Mao et al., 2007). It is the first mean of analyzing β-1,3–1,6-glucan with high precision, without extensive clean-up. The methods are useful tools to determine β-glucans in selected samples. In conclusion, the methods are useful tools to determine β-glucans in selected samples.

7. Biological properties of mushroom β-glucan

Mushrooms are also recognized as functional foods due to β-glucans that offer considerable beneficial impacts on human health. Brown and Gordon (2005) investigated all the known receptors for β-glucans and discussed the various immune responses initiated by β-glucan, with reference to fungal infection, in both vertebrates and invertebrates. Chen and Seviour (2007) discussed the mechanisms of action of these fungal β-glucans appear to depend on their capabilities to bind to cell receptors, which are known to include dectin-1, CR3, LacCer, and scavenger receptors. Chanput et al. (2012) examined different immunological aspects of β-glucans derived from different sources (shiitake, oat and barley) on phorbol myristate acetate differentiated THP-1 macrophages. Commercially purified barley β-glucan and lentinan were included to compare β-glucans from the same origin but different degree purity and processing. Based on *in vitro* analysis, it concluded that the analyzed β-glucans had varying levels of immuno-modulating properties, which were likely related to structure, molecular weight and compositional characteristic of β-glucan. Rop et al. (2009) presented a survey of the β-glucans found in Basidiomycetes and provide general information about their properties and beneficial effects on animal and human beings. Chen et al., 2013 summarized the use of mushroom β-glucans in colon cancer, and found that β-glucans could decrease the size of xenografted colon cancer tumours *via* the stimulation of the immune system and direct cytotoxicity. Moreover, lentinan, the backbone of β-(1,3)-glucan with β-(1,6) branches, is one of the active ingredients purified from mushroom Shiitake and has been approved as a biological response modifier for the treatment of gastric cancer in Japan (Ina et al., 2013). A growing body of science indicates that β-glucans promote health in a number of important ways, such as immuno-modulatory, antitumor, antiviral, (Borchers et al., 2004; Moradali et al., 2007), cardiovascular (Wasser and Weis, 1999), liver protective, anti-inflammatory, (Lindequist et al., 2005), radioprotective (Pillai and Devi, 2013), antidiabetic (Kim et al., 2005), antioxidant (Deng et al., 2012), antibacterial (Suay et al., 2000; Ishikawa et al., 2001; Shittu et al., 2005; Beattie et al., 2010), and antiobesity activities (Zhang et al., 2013). Antitumour activity (Kidd, 2000; Zhang et al., 2007; Deng et al., 2012; Ren et al., 2012) and immune-modulating activity (Wasser, 2002) of mushroom β-glucan have been documented in the previous reviews. Thus, the current review focuses on the antibacterial activity, antiviral activity and radioprotective effect of mushroom β-glucan. The biological activities of β-glucans from mushrooms are presented in Table 3.

7.1. Antibacterial activity

Several studies have suggested that mushroom β-glucan possesses antimicrobial activity. Highly branched glucan containing mainly 1,3- and 1,6-linkages from mushroom *Lentinula edodes* was tested for antibacterial activity *in vitro* against gram-positive and gram-negative bacteria. The minimum inhibitory concentration (MIC) values showed that this glucan had a potent antibacterial effect against different kind of bacteria such as *Bacillus megraterium* NCIB 2602 (MIC: 150 mg for sample), *Enterococcus phoeniculicola* JLB-1T (MIC: 150 mg for sample), *Klebsiella peneumoniae* K4 (MIC: 80 mg for sample) and so on (Rasmy et al., 2010). *S. commune* has been reported to produce exopolysaccharides (EPS), and secreted the β-glucans as a uniform, primary molecular structure. This EPS possesses a β-(1 → 3)-linked backbone with single β-(1 → 6)-linked glucose side chains, in which upon 100% oxidation will result in a large number of aldehyde groups that exhibit antibacterial properties (Jayakumar et al., 2010; Teon et al., 2012). However, the antimicrobial mechanism of glucan from mushroom *L. edodes* is still unclear. Matsuyama et al. (1992) evaluated the ability of β-glucans to enhance protection against bacterial infection in yellowtail. These β-glucans derived from mushroom *S. commune* and mushroom *S. glucanicum*. The results indicated that the β-1,3-glucans enhanced the resistance of yellowtail against bacterium *Streptococcus* spp. infection through the activation of the non-specific immune system.

Table 3
Biological activities of β-glucans from mushrooms.

Biological activities	Mushrooms β-glucans	Type of article	References
Immunological activity Antitumour activity	β-Glucans	Review	Borchers et al. (2004)
Antimicrobial activity	β-Glucans	Review	Lindequist et al. (2005) Soares et al. (2013)
Antiallergic activity Anti-inflammatory activity Antiatherogenic activity Hypoglycaemic activity Hepatoprotective activity			
Cardiovascular effect Hypercholesterolemia effect	β-Glucans	Review	Wasser and Weis (1999)
Antibacterial activity	β-Glucans	Original research	Beattie et al. (2010)
	β-Glucans	Original research	Ishikawa et al. (2001)
	β-Glucans	Original research	Shittu et al. (2005)
Radioprotective effect	β-Glucans	Original research	Pillai and Devi (2013)
		Patent	Ostroff and Ross (2005)
Antioxidant activity	β-Glucans	Original research	Deng et al. (2012)
Antidiabetic activity	β-Glucans	Original research	Kim et al. (2005)
	β-Glucans	Review	Chen and Raymond (2008)
Antiobesity activity	β-Glucans	Original research	Zhang et al. (2013)
Antiviral activity	β-Glucans	Review	Borchers et al. (2004)
		Original research	Minari et al. (2011)

7.2. Antiviral activity

Natural products are an inexhaustible source of compounds with promising pharmacological activities, including antiviral action. With respect to the antiviral potential, an extracted β-glucan from mushroom *A. brasiliensis* was investigated in the replication of bovine herpesvirus 1 (BoHV-1) in HepG-2 cell cultures (Minari et al., 2011). The β-glucan presented high inhibition of virus replication by plaque assay (83.2%) and immuno-fluorescence assay (63.8%). Although the mechanism is not clear yet, β-glucan is suggested to inhibit BoHV-1 replication by interfering with the early events of viral penetration.

7.3. Radioprotective effect

The *in vivo* radioprotective effect of mushroom β-glucan has been studied. Pillai and Devi (2013) investigated the protection effect offered by β-glucan from mushroom *G. lucidum* against radiation induced damage by taking mouse survival, haematology, liver glutathione, liver malondialdehyde and bone marrow chromosomal aberrations as markers. The radioprotective effect of β-glucan was compared with that of clinically used radioprotective drug amifostine, at 300 mg/kg body weight administered intraperitoneally, 30 min before irradiation. β-Glucan (500 μg/kg body weight) produced 66% mouse survival at 30 days given post irradiation, and 83% survived at 30 days with 300 mg/kg body weight of amifostine administered before radiation while radiation alone produced 100% mortality. Significant reduction in the number of aberrant cells and different types of aberration was observed in both β-glucan and amifostine administered groups compared to radiation alone treated group. Ostroff and Ross (2005) treated and prevented radiation and chemotherapy related injury or afflictions, such as myelosuppression and decreased macrophage activity, by administering a prophylactically or therapeutically effective amount of particulate, bioavailable β-(1,3–1,6) glucan as a radioprotectant.

8. Conclusions

Mushrooms have been playing important roles in several aspects of human activity. Due to these properties, mushrooms have been recognized as functional foods, and as a source for the development of medicines, nutraceuticals, and cosmeceuticals. Mushroom β-glucan is an effective and ideal component for infusing immune health benefits into any kind of foods, baked goods, beverage and dietary supplements. On one hand, extraction and purification of β-glucan from mushroom is a difficult job. The challenge now exists to optimize extraction and purification procedures of β-glucan. On the other hand, the biological activities of β-glucans derived from edible and medicinal mushrooms have received much attention in biomedical sciences. However, the mechanism of the biological activity of β-glucans isolated from edible and medicinal mushrooms is still not completely understood. To further improve the possible utilization of β-glucan from mushroom, future research should be focused on in the following directions: (1) a better definition of chemical structure–biological properties relationship; (2) product development methods to develop foods with β-glucan as functional ingredients; (3) possible research directions including atomic force microscopy of structure and visualization of super-helix structure to better illustrate the structure of β-glucan.

Conflict of interest

None.

Compliance with ethics requirements

This article does not contain any studies with human or animal subjects.

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References

AACC, 2011. AACC International Approved Methods 32-23.01. Beta-glucan Content of Barley and Oats – Rapid Enzymatic Procedure. American Association of Cereal Chemists, St Paul, MN.

Ahmad, A., Anjum, F.M., Zahoor, T., Nawaz, H., Dilshad, S.M.R., 2012. Beta glucan: a valuable functional ingredient in foods. Critical Reviews in Food Science and Nutrition 52, 201–212.

Alves, M.J., Ferreira, I.C.F.R., Dias, J., Teixeira, V., Martins, A., Pintado, M., 2012. A review on antimicrobial activity of mushroom (*Basidiomycetes*) extracts and isolated compounds. Planta Medica 78, 1707–1718.

Andersson, K., Svedberg, K., Lindholmb, M., Ostec, R., Hellstranda, P., 2010. Oats (*Avena sativa*) reduce atherogenesis in LDL-receptor-deficient mice. Atherosclerosis 212, 93–99.

AOAC, 1995. AOAC Official Method 995.16. β-D-Glucan in Barley and Oats Streamlined Enzymatic Method. First Action. AOAC.

Bae, I., Lee, S., Kim, S., Lee, H., 2009. Effect of partially hydrolyzed oat β-glucan on the weight gain and lipid profile of mice. Food Hydrocolloids 23, 2016–2021.

Beattie, K.D., Rouf, R., Gander, L., May, T.W., Ratkowsky, D., Donner, C.D., et al., 2010. Antibacterial metabolites from Australian macrofungi from the genus *Cortinarius*. Phytochemistry 71, 948–955.

Borchers, A., Keen, C.L., Gershwin, M.E., 2004. Mushrooms, tumors, and immunity: an update. Experimental Biology and Medicine 229, 393–406.

Brennan, C.S., Cleary, L.J., 2005. The potential use of cereal (1 → 3, 1 → 4)-β-D-glucans as functional food ingredients. Journal of Cereal Science 42, 1–13.

Brown, G.D., Gordon, S., 2005. Immune recognition of fungal β-glucans. Cellular Microbiology 7, 471–479.

Buckeridge, M.S., Rayon, C., Urbanowicz, B., Tiné, M.A.S., Carpita, N.C., 2004. Mixed linkage (1 → 3), (1 → 4)-β-D-glucans of grasses. Cereal Chemistry 81, 115–127.

Camellini, C.M., Maraskin, M., De Mendonca, M.M., Zucco, C., Ferreira, A.G., Tavares, L.A., 2005. Structural characterization of beta-glucans of *Agaricus brasiliensis* in different stages of fruiting body maturity and their use in nutraceutical products. Biotechnology Letters 27, 1295–1299.

Carbonero, E.R., Ruthes, A.C., Freitas, C.S., Utrilla, P., Gálvez, J., da Silva, E.V., et al., 2012. Chemical and biological properties of a highly branched β-glucan from edible mushroom *Pleurotus sajor-caju*. Carbohydrate Polymers 90, 814–819.

Cha, Y.J., Alam, N., Lee, J.S., Lee, K.R., Shim, M.J., Lee, M.W., et al., 2012. Anticancer and immunopotentiating activities of crude polysaccharides from *Pleurotus nebrodensis* on mouse sarcoma 180. Mycobiology 40, 236–243.

Chanput, W., Reitsma, M., Kleinjans, L., Mes, J.J., Savelkoul, H.F., Wichers, H.J., 2012. β-Glucans are involved in immune-modulation of THP-1 macrophages. Molecular Nutrition & Food Research 56, 822–833.

Chen, J.Z., Raymond, K., 2008. Beta-glucans in the treatment of diabetes and associated cardiovascular risks. Journal of Vascular Health and Risk Management 4, 1265–1272.

Chen, J.Z., Seviour, R., 2007. Medicinal importance of fungal β-(1 → 3) (1 → 6)-glucans. Mycological Research 111, 635–652.

Chen, J., Zhang, X.D., Jiang, Z., 2013. The application of fungal beta-glucans for the treatment of colon cancer. Anti-Cancer Agents in Medicinal Chemistry 13, 725–730.

Chihara, G., 2001. International Journal of Oriental Medicine. Oriental Healing Arts Institute of U.S.A Press, , pp. 261–266.

Daou, C., Zhang, H., 2012. Oat beta-glucan: its role in health promotion and prevention of diseases. Comprehensive Reviews in Food Science and Food Safety 11, 355–365.

Das, D., Mondal, S., Roy, S.K., Maiti, D., Bhunia, B., Maiti, T.K., et al., 2010. A (1 → 6)-beta-glucan from a somatic hybrid of *Pleurotus florida* and *Volvariella volvacea*: isolation, characterization and study of immunoenhancing properties. Carbohydrate Research 345, 974–978.

Deng, C., Hu, Z., Fu, H.T., Hu, M.H., Xu, X., Chen, J.H., 2012. Chemical analysis and antioxidant activity *in vitro* of a β-D-glucan isolated from *Dictyophora indusiata*. International Journal of Biological Macromolecules 51, 70–75.

Dong, Q., Wang, Y., Shi, L., Yao, J., Li, J., Ma, F., et al., 2012. A novel water-soluble β-D-glucan isolated from the spores of *Ganoderma lucidum*. Carbohydrate Research 353, 100–105.

Du, B., Bian, Z.X., Xu, B.J., 2013. Skin health promotion effects of natural beta-glucan derived from cereals and microorganisms: a review. Phytotherapy Research, http://dx.doi.org/10.1002/ptr.4963 (in press).

Fang, J., Wang, Y., Lv, X., Shen, X., Ni, X., Ding, K., 2012. Structure of a β-glucan from *Grifola frondosa* and its antitumor effect by activating Dectin-1/Syk/NF-κB signaling. Glycoconjugate Journal 29, 365–377.

Guerra Dore, C.M., Azevedo, T.C., de Souza, M.C., Rego, L.A., de Dantas, J.C., Silva, F.R., et al., 2007. Antiinflammatory antioxidant and cytotoxic actions of β-glucan-rich extract from *Geastrum saccatum* mushroom. International Immunopharmacology 7, 1160–1169.

Handayani, D., Meyer, B.J., Chen, J.Z., Tang, P., Kwok, P.C.L., Chan, H.K., et al., 2012. The comparison of the effect of oat and shiitake mushroom powder to prevent body weight gain in rats fed high fat diet. Food and Nutrition Sciences 3, 1009–1019.

Havrlentová, M., Petruláková, Z., Burgárová, A., Gago, F., Hlinková, A., Sturdík, E., 2011. Cereal β-glucans and their significance for the preparation of functional foods – a review. Czech Journal of Food Sciences 29, 1–14.

Huang, X., Tu, Z., Jiang, Y., Xiao, H., Zhang, Q., Wang, H., 2012. Dynamic high pressure microfluidization-assisted extraction and antioxidant activities of lentinan. International Journal of Biological Macromolecules 51, 926–932.

Ina, K., Kataoka, T., Ando, T., 2013. The use of lentinan for treating gastric cancer. Anti-Cancer Agents in Medicinal Chemistry 13, 681–688.

Ishikawa, N.K., Kasuya, M.C.M., Vanetti, M.C.D., 2001. Antibacterial activity of *Lentinula edodes* grown in liquid medium. Brazilian Journal of Microbiology 32, 206–210.

Jayakumar, G.C., Kanth, S.V., Chandrasekaran, B., Rao, R.J., Nair, B.U., 2010. Preparation and antimicrobial activity of scleraldehyde from *Schizophyllum commune*. Carbohydrate Research 345, 2213–2219.

Jin, Y., Zhang, L., Chen, L., Chen, Y., Cheung, P.C., Chen, L., 2003. Effect of culture media on the chemical and physical characteristics of polysaccharides isolated from *Poria cocos* mycelia. Carbohydrate Research 338, 1507–1515.

Kalac, P., 2009. Chemical composition and nutritional value of European species of wild growing mushrooms: a review. Food Chemistry 113, 9–16.

Kalac, P., 2013. A review of chemical composition and nutritional value of wild-growing and cultivated mushrooms. Journal of the Science of Food and Agriculture 93, 209–218.

Kang, S.C., Koo, H.J., Park, S., Lim, J.D., Kim, Y.J., Kim, T., et al., 2013. Effects of β-glucans from *Coriolus versicolor* on macrophage phagocytosis are related to the Akt and CK2/Ikaros. International Journal of Biological Macromolecules 57, 9–16.

Kao, P.F., Wang, S.H., Hung, W.T., Liao, Y.H., Lin, C.M., Yang, W.B., 2012. Structural characterization and antioxidative activity of low-molecular-weights beta-1,3-glucan from the residue of extracted *Ganoderma lucidum* fruiting bodies. Journal of Biomedicine and Biotechnology 2012, 1–8.

Kerckhoffs, D., Hornstra, G., Mensink, R., 2003. Cholesterol-lowering effect of beta-glucan from oat bran in mildly hypercholesterolemic subjects may decrease when beta-glucan is incorporated into bread and cookies. American Journal of Clinical Nutrition 78, 221–227.

Kidd, P.M., 2000. The use of mushroom glucans and proteoglycans in cancer treatment. Alternative Medicine Review 5, 4–27.

Kim, Y.W., Kim, K.H., Choi, H.J., Lee, D.S., 2005. Anti-diabetic activity of β-glucans and their enzymatically hydrolyzed oligosaccharides from *Agaricus blazei*. Biotechnology Letters 27, 483–487.

Klaus, A., Kozarski, M., Niksic, M., Jakovljevic, D., Todorovic, N., van Griensven, L.J.L.D., 2011. Antioxidative activities and chemical characterization of polysaccharides extracted from the Basidiomycete *Schizophyllum commune*. LWT – Food Science and Technology 44, 2005–2011.

Ko, Y.T., Lin, Y.L., 2004. 1,3-Beta-glucan quantification by a fluorescence microassay and analysis of its distribution in foods. Journal of Agricultural and Food Chemistry 52, 3313–3318.

Kumari, M., Survase, S.A., Singhal, R.S., 2008. Production of schizophyllan using *Schizophyllum commune* NRCM. Bioresource Technology 99, 1036–1043.

Kyanko, M.V., Canel, R.S., Ludemann, V., Pose, G., Wagner, J.R., 2013. β-Glucan content and hydration properties of filamentous fungi. Applied Biochemistry and Microbiology 49, 41–45.

Lai, M.N., Ng, L.T., 2013. Antioxidant and antiedema properties of solid-state cultured honey mushroom, *Armillaria mellea* (higher Basidiomycetes) extracts and their polysaccharide and polyphenol contents. International Journal of Medicinal Mushrooms 15, 1–8.

Laroche, C., Michaud, P., 2007. New developments and prospective applications for β-(1,3) glucans. Recent Patents on Biotechnology 1, 59–73.

Lazaridou, A., Biliaderis, C.G., 2007. Molecular aspects of cereal β-glucan functionality: physical properties technological applications and physiological effects. Journal of Cereal Science 46, 101–118.

Lindequist, U., Niedermeyer, T.H.J., Jülich, W.D., 2005. The pharmacological potential of mushrooms. Evidence-Based Complementary and Alternative Medicine 2, 285–299.

Ma, Z., Wang, J., Zhang, L., 2008. Structure and chain conformation of beta-glucan isolated from *Auricularia auricula-judae*. Biopolymers 89, 614–622.

Maity, K.K., Patra, S., Dey, B., Bhunia, S.K., Mandal, S., Bahera, B., Maiti, T.K., Sikdar, S.R., Islam, S.S., 2013. A β-glucan from the alkaline extract of a somatic hybrid (PfloVv5FB) of *Pleurotus florida* and *Volvariella volvacea*: structural characterization and study of immunoactivation. Carbohydrate Research 370, 13–18.

Manzi, P., Pizzoferrato, L., 2000. Beta-glucans in edible mushrooms. Food Chemistry 68, 315–318.

Mao, C.F., Hsu, M.C., Hwang, W.H., 2007. Physicochemical characterization of grifolan: thixotropic properties and complex formation with Congo Red. Carbohydrate Polymers 68, 502–510.

Masuda, Y., Togo, T., Mizuno, S., Konishi, M., Nanba, H., 2012. Soluble β-glucan from *Grifola frondosa* induces proliferation and Dectin-1/Syk signaling in resident macrophages via the GM-CSF autocrine pathway. Journal of Leukocyte Biology 91, 547–556.

Matsuyama, H., Mangindaan, R.E.P., Yano, T., 1992. Protective effect of schizophyllan and scleroglucan against *Streptococcus* sp infection in yellowtail (*Seriola quinqueradiata*). Aquaculture 101, 197–203.

Mattila, P., Suonpaa, K., Piironen, V., 2000. Functional properties of edible mushrooms. Nutrition Reviews 16, 694–696.

McCleary, B.V., Codd, R., 1991. Measurement of (1-3)(1-4)-β-D-glucan in barley and oats: a streamlined enzymatic procedure. Journal of the Science of Food and Agriculture 55, 303–312.

Minari, M.C., Rincão, V.P., Soares, S.A., Ricardo, N.M., Nozawa, C., Linhares, R.E., 2011. Antiviral properties of polysaccharides from *Agaricus brasiliensis* in the replication of bovine herpesvirus 1. Acta Virologica 55, 255–259.

Mizuno, T., Minato, K., Kawakami, S., Tatusuoka, S., Denpo, Y., Tsuchida, H., 2001. Contents of anti-tumor polysaccharides in certain mushrooms and their immunomodulating activities. Food Science and Technology Research 7, 31–34.

Moradali, M.F., Mostafavi, H., Ghods, S., Hedjaroude, G.A., 2007. Immunomodulating and anticancer agents in the realm of macromycetes fungi (macrofungi). International Immunopharmacology 7, 701–724.

Nitschke, J., Modick, H., Busch, E., von Rekowski, R.W., Altenbach, H.J., Mölleken, H., 2011. A new colorimetric method to quantify β-1,3–1,6-glucans in comparison with total β-1,3-glucans in edible mushrooms. Food Chemistry 127, 791–796.

Ostroff, G.R., Ross, G.D., 2005. Methods of using beta glucan as a radioprotective agent. US patent 20050245480 A1.

Petravic-Tominac, V., Zechner-Krpan, V., Grba, S., Srecec, S., Panjkota-Krbavcic, I., Vidovic, L., 2010. Biological effects of yeast β-glucans. Agriculturae Conspectus Scientificus 75, 149–158.

Pillai, T.G., Devi, U.P., 2013. Mushroom beta glucan: potential candidate for post irradiation protection. Mutation Research 751, 109–115.

Poucheret, P., Fons, F., Rapior, S., 2006. Biological and pharmacological activity of higher fungi: 20-Year retrospective analysis. Cryptogamie Mycologie 27, 311–333.

Rasmy, G.E., Botros, W.A., Kabeil, S., Daba, A.S., 2010. Preparation of glucan from *Lentinula edodes* edible mushroom and elucidation of its medicinal value. Australian Journal of Basic and Applied Sciences 4, 5717–5726.

Ren, L., Perera, C., Hemar, Y., 2012. Antitumor activity of mushroom polysaccharides: a review. Food & Function 3, 1118–1130.

Rieder, A., Samuelsen, A.B., 2012. Do cereal mixed-linked β-glucans possess immune-modulating activities? Molecular Nutrition & Food Research 56, 536–547.

Rop, O., Mlcek, J., Jurikova, T., 2009. Beta-glucans in higher fungi and their health effects. Nutrition Reviews 67, 624–631.

Satitmanwiwat, S., Ratanakhanokchai, K., Laohakunjit, N., Chao, L.K., Chen, S.T., Pason, P., et al., 2012. Improved purity and immunostimulatory activity of β-(1 → 3)(1 → 6)-glucan from *Pleurotus sajor-caju* using cell wall-degrading enzymes. Journal of Agricultural and Food Chemistry 60, 5423–5430.

Schmid, F., Stone, B.A., McDougall, B.M., Bacic, A., Martin, K.L., Brownlee, R.T., et al., 2001. Structure of epiglucan a highly side-chain/branched (1 → 3;1 → 6)-β-glucan from the micro fungus *Epicoccum nigrum* Ehrenb. ex Schlecht. Carbohydrate Research 331, 163–171.

Sen, I.K., Maji, P.K., Behera, B., Maiti, T.K., Mallick, P., Sikdar, S.R., et al., 2013. Glucan of a somatic hybrid mushroom pfls1h: structural characterization and study of immunological activities. International Journal of Biological Macromolecules 53, 127–132.

Shibata, A., Hida, T.H., Ishibashi, K., Miura, N.N., Adachi, Y., Ohno, N., 2012. Disruption of actin cytoskeleton enhanced cytokine synthesis of splenocytes stimulated with beta-glucan from the cauliflower medicinal mushroom, *Sparassis crispa* Wulf.:Fr (higher Basidiomycetes) *in vitro*. International Journal of Medicinal Mushrooms 14, 257–269.

Shittu, O.B., Alofe, F.V., Onawunmi, G.O., Ogundaini, A.O., Tiwalade, T.A., 2005. Mycelial growth and antibacterial metabolite production by wild mushrooms. African Journal of Biomedical Research 8, 157–162.

Smiderle, F.R., Carbonero, E.R., Mellinger, C.G., Sassaki, G.L., Gorin, P.A., Iacomini, M., 2006. Structural characterization of a polysaccharide and a beta-glucan isolated from the edible mushroom *Flammulina velutipes*. Phytochemistry 67, 2189–2196.

Soares, A.A., de Sá-Nakanishi, A.B., Bracht, A., da Costa, S.M., Koehnlein, E.A., de Souza, C.G., et al., 2013. Hepatoprotective effects of mushrooms. Molecules 18, 7609–7630.

Stephanie, P.D., Fatma, K., Trevor, J.M., Marcos, M.A., Alan, A.H., Rafael, K., 2007. Probing size exclusion mechanisms of complex hydrocarbon mixtures: the effect of altering eluent compositions. Energy and Fuels 21, 3484–3489.

Suay, I., Arenal, F., Asensio, F.J., Basilio, A., Cabello, M.A., Díez, M.T., et al., 2000. Screening of basidiomycetes for antimicrobial activities. Antonie van Leeuwenhoek 78, 129–139.

Synytsya, A., Novak, M., 2013. Structural diversity of fungal glucans. Carbohydrate Polymers 92, 792–809.

Tao, Y.Z., Zhang, L.N., Cheung, P.C.K., 2006. Physicochemical properties and antitumor activities of water-soluble native and sulfated hyperbranched mushroom polysaccharides. Carbohydrate Research 341, 2261–2269.

Teon, Y.P., Don, M.M., Ujang, S., 2012. Nutrient improvement using statistical optimization for growth of *Schizophyllum commune* and its antifungal activity against wood degrading fungi of rubberwood. Biotechnology Progress 28, 232–241.

Villares, A., Mateo-Vivaracho, L., Guillamon, E., 2012. Structural features and healthy properties of polysaccharides occurring in mushrooms. Agriculture 2, 452–471.

Wang, Y., Zhang, L., Li, Y., Hou, X., Zeng, F., 2004. Correlation of structure to antitumor activities of five derivatives of a beta-glucan from *Poria cocos* sclerotium. Carbohydrate Research 339, 2567–2574.

Wang, J.G., Ma, Z.C., Zhang, L.N., Fang, Y.P., Jiang, F.T., Phillips, G.O., 2011. Structure and chain conformation of water-soluble heteropolysaccharides from *Ganoderma lucidum*. Carbohydrate Polymers 86, 844–851.

Wasser, S.P., 2002. Medicinal mushrooms as a source of antitumor and immunomodulating polysaccharides. Applied Microbiology and Biotechnology 60, 258–274.

Wasser, S.P., Weis, A.L., 1999. Medicinal properties of substances occurring in higher Basidiomycetes mushrooms: current perspectives (review). International Journal of Medicinal Mushrooms 1, 31–62.

Wood, P.J., 2007. Cereal β-glucans in diet and health. Journal of Cereal Science 46, 230–238.

Wood, P.J., Beer, M.U., Butler, G., 2000. Evaluation of the role of concentration and molecular weight of oat β-glucan in determining effect of viscosity on plasma glucose and insulin following an oral glucose load. British Journal of Nutrition 84, 19–23.

Xu, S., Xu, X., Zhang, L., 2012. Branching structure and chain conformation of water-soluble glucan extracted from *Auricularia auricula-judae*. Journal of Agricultural and Food Chemistry 60, 3498–3506.

Zekovic, D.B., Kwiatkowski, S., Vrvic, M.M., Jakovljevic, D., Moran, C.A., 2005. Natural modified $(1 \rightarrow 3)$-β-glucans in health promotion and disease alleviation. Critical Reviews in Biotechnology 25, 205–230.

Zhang, M., Cheung, P.C.K., Zhang, L., 2001. Evaluation of mushroom dietary fiber (nonstarch polysaccharides) from sclerotia of *Pleurotus tuber-regium* (Fries) Singer as a potential antitumor agent. Journal of Agricultural and Food Chemistry 49, 5059–5062.

Zhang, M., Cui, S.W., Cheung, P.C.K., Wang, Q., 2007. Antitumor polysaccharides from mushrooms: a review on their isolation process structural characteristics and antitumor activity. Trends in Food Science & Technology 18, 4–19.

Zhang, Y., Xia, L., Pang, W., Wang, T., Chen, P., Zhu, B., et al., 2013. A novel soluble β-1,3-D-glucan salecan reduces adiposity and improves glucose tolerance in high-fat diet-fed mice. British Journal of Nutrition 109, 254–262.