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Fascinating Vital Mushrooms. Tinder Fungus (*Fomes fomentarius* (L.) Fr.) as a Dietary Supplement

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ABSTRACT

A composition of the fruiting bodies of Fomes fomentarius and their extracted fibers were studied. The cell walls of the fungus composed mainly of glucan, chitin and melanin-like substances with hemicellulose, uronic and glucuronic acids as minor compounds. In comparison with the raw fruiting bodies, the fibres from Fomes fomentarius, extracted under mild conditions, contain about one third of fats, half of alpha-glucans, 25% less melanin-like substances, 15% less hemicellulose, around 25% more beta-glucans and 15% more chitin. While under severe extraction conditions a gradually increase of the chitin percentage in the cell walls is possible, the eco-friendly purification method is preferable. After mild hot purification, the fibres could be more bioavailable and accessible for specific receptors, immune cells, intestinal microbiota etc.to be a good candidate to replenish a number of vital mushrooms used as dietary supplements.

Keywords: *Tinder fungus, Fomes fomentarius, composition, vital mushrooms, dietary supplement*

INTRODUCTION

Fungi are very special, fascinating organisms. Most of them contain biologically active substance. Nowadays, the pharmacological effects of around 700 fungal species are known [1]. They can e.g. hinder growth of various tumours, stimulate the immune system, support cell regeneration, help rid the body of toxins, support the well being etc. [1-4].

As a result of the increased interest in vital fungi and their positive characteristics, rather unknown or forgotten types are being examined today more closely. The tinder fungus, also called amadou mushroom, (*Fomes fomentarius* (L.) Fr.) is among those fungi (Fig. 1**)**.

Figure1. *A fruiting body of thetinder fungus (Fomes fomentarius (L.)Fr.)*

Ages ago the middle layer of the fruiting bodies of the *Fomes fomentarius* were used as tinder or to create a leather-like material. Because of its positive attributes, tinder fungus has also been used in folk medicine for treatment of gastro-intestinal, liver-related problems and inflammations, for pain relief, as a hemostatic and anticancer remedy etc. [5-9].

In Europe, the first wave of scientific interest in the tinder fungus arrived since the 1970s, again with examinations of the composition of the fungal extracts [10]. Literature research shows, that interest in the topic has not vanished until today, but on the contrary, is growing.

Nowadays, many scientific examinations and clinical studies point out, that the isolated cell wall and extracts of *Fomes fomentarius* can positively influence the immune system [11-13], blood sugar levels, cholesterol levels [14] and also have an anti-bacterial, anti-viral, fungicidal, anti-inflammatory, and analgesic effects [11, 15-18]. They can also absorb infections in the gastro-intestinal area [11], show anti-cancer activities [5, 19-21] and bind heavy metals, radionuclides, and free radicals [6, 11, 22-30]. Because of their positive attributes, tinder fungus products are merchandised as dietary supplements.

In this paper we will focus on the analysis of the milled fruit bodies and insoluble extracted fibres.

MATERIAL AND METHODS

Identification of Species

Fomes fomentarius (L.) Fr.) fruiting bodies were identified morphologically and using DNA-analysis. DNA-analysis was performed by Alvalab molecular analysis service, LA Rochela, Spain, using PCR and sequencing of parts of the ITS-region [31].

Extraction of the Fibres

Extraction/purification comprises the following steps: drying of the fruiting bodies, grinding, mixing with extracting liquid, and extraction at high temperature, washing to cleanse the final product from residual extracting agent and soluble components, and drying of the final product.

After a mild purification the extracted insoluble fibrous cell walls (extracted fibers) were obtained (also known as Good Feeling Power®) [32].

Severe modification of the cell walls (chitin-modificated fibres) was possible as a result of repetitive treatment with 5-20% acidic (H_2SO_4, HCl) and basic (NaOH, Na_2CO_3) solutions.

Dietary Fibres

Total dietary fibres that are primarily hemicelluloses, pectins, other non-starch hydrocolloids, resistant starch, cellulose and lignin were estimated gravimetrically after extraction and enzymatic digestion of non-fibre material according to ICC standard method No. 156 [33].

Lipid Fraction

Crude fat was determined as a sum of wax, resins, lipids and fat according to NREL/TP-510-42619 (2008) [34].

Detection of Protein Content

Proteinogenic amino acids were measured after hydrolysis using HPLC with a fluorescence detector [35].

The AOAC-approved method of Kjeldahl was performed to measure total nitrogen and to determine total protein content [36].

For protein calculation, the standard factor of 4.16 was use as recommend for fungi containing non-protein nitrogen [37, 38].

Determination of Sugars

The sugars glucose, fructose, sucrose, lactose and maltose were extracted with demineralized water (60°C) from the sample matrix. After injection to the ion chromatographic system the sugars were separated by means of an anion exchange column (CarboPac PA 20, 3x150 mm) and sodium hydroxide as eluent (concentration 65 mmol, temperature 30°C, flow 0,4 ml/min). The separated sugars were detected electrochemically by means of the integrated pulsed amperometry with a gold electrode. The quantification was made with a calibration by means of external standard solutions in different concentrations.

Detection of Chitin, Glucan, Hemicellulose, Glucuronic and Uronic Acids

Acid hydrolysis according to NREL/TP-510-42618 followed by High-performance Anion Exchange Chromatography coupled with Pulsed Amperometric Detection (HPAE-PAD) was used to estimate Glucan, Hemicellulose, Glucuronic and Uronic acids [39].

The chitin amount was estimated after hydrolysis with HCl followed by HPAE-PAD [40] or by colorimetric modified method of Chen & Johnson [41]. A drying step in the method was replaced with titration of the acid solution till neutral pH 7.

The total-, alpha- and beta-glucans were determined using an enzyme based test kit developed for mushrooms and yeasts (Megazyme Ltd., Wicklow, Ireland) according to the manufacturer's instructions [42].

Melanin-Like Insoluble Residue

Melanin-like insoluble substances were estimated as residue after acid hydrolysis of the samples according to NREL/TP-510-42618 [39].

RESULTS AND DISCUSSION

Microscoping of isolated fibres shows that the cell walls of the tinder fungus have well-formed fibrous structure composed of long cell walls that are empty inside (Fig. 2A). The fibres have a cell wall thickness of 0.2-1 µm, a diameter of 3-5 µm, and a length of up to one millimeter (Fig. 2B) [32].

The fibers isolated from tinder fungus fruit bodies are composed of a number of polymers (Table 1) which are bound together to form a three-dimensional chemically and mechanically stable netlike structure.

Upon the comparative analysis of the glucosamine and acetyl residues in acid hydrolysate (HPAE-PAD and HPLC-RI, respectively), chitin in the fibres will be approximately 72% deacetylated.

Hydrolysis residue, presented in Table 1, is a complex acid insoluble phenolic biopolymer. This polymer can include insoluble lignin, acid insoluble proteins and melanins. The tinder fungus mostly nourishes itself from brown lignin in wood, leaving behind the white colored celluloses. This is how the fungus creates the so-called "white rot" in wood [43]. Due to relative high amount of nitrogen (around 10%), the hydrolysis residues in the study were quantified as melanin-like substances.

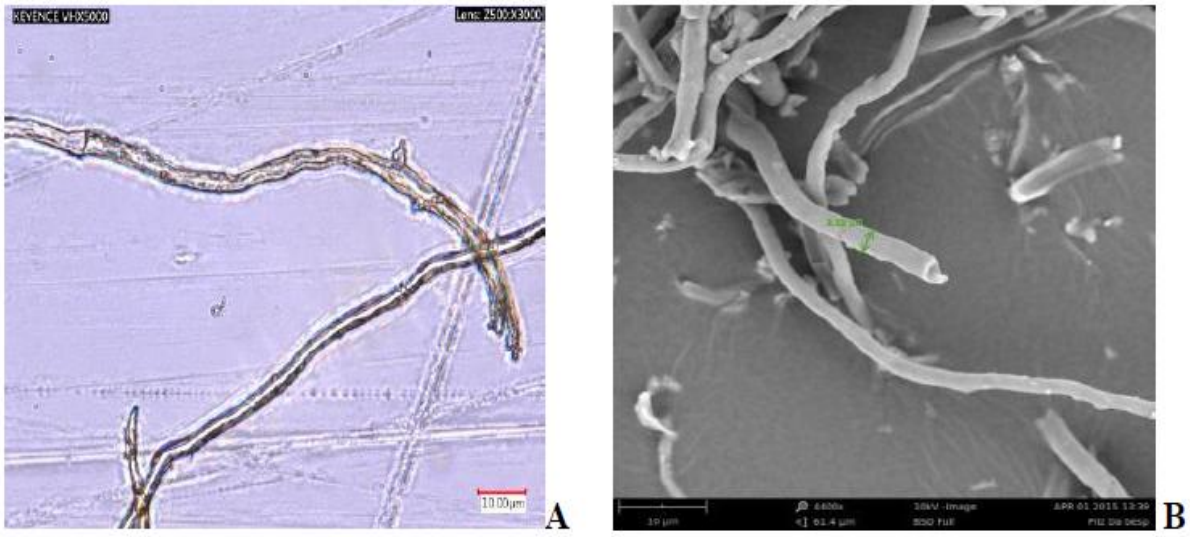

Figure2. *The fibres isolated from tinder fungus fruit bodies. A - Keyence VHX-5000 digital microscope, KEYENCE DEUTSCHLAND GmbH, 3000x. B - The Phenom ProX scanning electron microscope (SEM), LOT-QuantumDesign GmbH, 4400x*

Melanins are a large diverse group of hydrophobic, negatively charged macromolecules formed by oxidative polymerization of phenolic or indolic compounds. Data of the chemical structure of fungal melanin pigments are very limited due to their heterogeneity, stability and complicated polymeric structure.

There are several different types of melanin, but DHN-melanin (named for one of the pathway intermediates, 1,8-dihydroxynaphthalene) and DOPA-melanin (named for one of the precursors, L-3,4-dihydroxyphenylalanine) have been proposed as the most important types in fungi [44, 45].

The brown pigments from *Fomes fomentarius* were detected also as melanins using nanosecond laser technology developed by LTB Lasertechnik Berlin GmbH and Magnosco GmbH [46]. Analytically, the three main structural components of the fibres (glucan, chitin and melanin-like substances) as well as the minor compounds (hemicellulose as a sum of mannan, galactan, xylan, arabinan and rhamnan, as well uronic and glucuronic acids) are detected as dietary fibres (Table 1).

In comparison with raw fruiting bodies, the extracted fibres contained about one third of ethanol extractives (fats, waxes, lipids, resins etc.), 25% less melanin-like substances, 15% less hemicellulose, but 15% more chitin.

Thus, the composition of the cell walls was influenced by removing the easy extractable soluble compounds.

Table1. *Composition of main copolymers building the cell walls of Fomes fomentarius in extracted fibres. The average data presented in g per 100 g of dry mass±SD*

Polymers	Mean, g/100 g dm	±SD	% to raw fruiting bodies
Chitin	6.70	0.23	117.5
Glucan	38.46	2.24	129.5
Hydrolysis residue (melanins)	22.16	3.10	75.9
Uronic acid	1.35	0.09	79.4
Glucuronic acid	1.33	0.12	121.2
Dietary fibres	74.93	1.07	-
Hemicellulose:	1.40	0.25	82.6

Mannan	0.66	0.12	-
Galactan	0.22	0.14	-
Xylan	0.55	0.27	-
Arabinan	0.11	0.02	-
Rhamnan	0.02	0.02	-

To qualify the total glucans, a Megazyme kit was used. The assay was previously described and is expected to deliver reliable results for glucans, and especially 1,3/1,6-ß-D-glucan content, in mushrooms and mushroom preparations [42, 47-51].

According to the data, presented in Table 2, the glucan in the fibres extracted from the *Fomes fomentarius* composed mainly of beta-glucans with less than 4% of alpha-glucans. Indeed, in the raw material the percentage of the alpha-glucans was around 7-10% of the total glucans (Table 2) [50].

Thus, in extracted fibres the amount of alpha-glucans decreases by the factor 2.

After mild proceeding of the raw material the amount of total- and beta-glucans slightly increased (Table 2).

Earlier published data [50] also show the lower amount of total glucans (24.94±1.832) and beta-glucans (22.495±2.329) in milled fruit bodies of *Fomes fomentarius*.

Table2. *Glucan content in the Fomes fomentarius milled fruiting bodies (raw material) and extracted fibres was detected using Megazyme assay. The average data presented in g per 100 g of dry mass±SD*

Fomes fomentarius	**Total glucans, g/100 g dm**	**±SD**	**Alpha-glucans, g/100 g dm**	**±SD**	**Beta-glucans, g/100 g dm**	**±SD**
Raw material	31.930	3.189	2.359	0.236	29.571	3.425
Extracted fibres	40.77	6.36	1.28	0.14	39.49	6.33
% glucans, in extracted fibres to raw material	127.69	-	54.31	-	133.55	-

Thus, glucan extracted from the cell wall of *Fomes fomentarius* belong mostly to the insoluble, highly molecular 1,3/1,6-ß-D-glucans, which show the highest bioactivity [52, 53].

Alone with molecular weight, the conformation of the glucans like single helix, double or triple helix is important.

To date, single helix or slightly loosened up triple helix lead to elevated macrophage activation [54-57]. Thus, purification of the fibers from the waxes, lipids and soluble cell compounds could lead to activation of the polymers.

To date there are several approaches to obtain fibres from the mushroom *Fomes fomentarius*. It could be isolated by repetitive extraction with different solvents (water, alcohol, benzene, alkali, acid).

After those severe extractions a product still has a fibrous structure and altered chitin-glucan-melanin distribution [58, 59].

A modification of the cell walls is possible as a result of repetitive treatment with acidic and basic solutions. One of the possible variations of the chitin percentage in the cell walls of the tinder fungus is presented in the table 3.

During modification experiments the chitin amount was estimated colorimetrically according to a modified method [41].

For the fibres extracted under mild conditions (Good Feeling Power®, table 3), the results are comparable with the HPAE-PAD methods (7.97% chitin vs. 6.7%, respectively) (Tables 1 and 3). Thus, the chitin amount can be estimated routinely using nonexpensive colorimetrical method.

According to our experiments, a modification of the tinder fungus fibres is rather possible. But using the eco-friendly mild purification method is preferable due to the lower amount of chemical waste.

Table3. *Increasing of the glucan percentage in cell walls of the Fomes fomentarius after repetitive treatment with acidic (HCl) and basic (NaOH) solutions (chitin-modificated fibres) in comparison with fibres extracted under mild conditions (Good Feeling Power®). The average data presented in g per 100 g of dry mass±SD*

Fomes fomentarius	**Chitin, g/100 g dm**
Chitin-modificated fibres	55.38±3.93
Good Feeling Power®	7.97±0.52

Protein Composition

The total protein content in tinder fungus fibres is estimated by HPLC as the total amino acid content after hydrolysis (Table 4) as well as using nitrogen content measurement (Kjeldahl assay).

Table 4. *Protein fraction after hydrolysis in tinder fungus fibres. The average data presented in g per kg of dry mass±SD*

Amino acids	Mean, g/kg dm	±SD
Aspartic acid	2.7	0.5
Glutamic acid	2.2	0.4
Serine	1.4	0.3
Histidine	0.3	0.1
Glycine	1.6	0.2
Threonine	1.4	0.3
Arginine	0.5	0.1
Alanine	1.5	0.2
Tyrosine	0.6	0.1
Valine	1.5	0.2
Phenylalanine	1.1	0.2
Isoleucine	1.2	0.2
Leucine	1.9	0.3
Lysine	0.4	0.0
Total	18.2	2.8

After hydrolysis the total amino acid content represents 1.8 % of extracted fibres. Using Kjeldahl assay and mushroom protein correlation factor 4.16 [37, 38], the protein amount in *Fomes fomentarius* extracted fibres was 4.49%±0.03.

This value seems low, but nevertheless is 2.5 times higher as observed after hydrolysis and HPLC analysis.

Thus, using conventional methods based on nitrogen estimation, misleading information can be obtained due to the presence of amid groups of chitin and melanin which interferes with these assays.

The conversion factor of total nitrogen into crude protein could vary significantly 3.45 - 4.38 [38, 60].

A comparable low level of proteins after hydrolysis (3.3%) was published for extracted fibres from *Aspergillus niger* [61].

Lipid Fraction

The fibres extracted from *Fomes fomentarius* contains ≤1.9% of crude fat (Table 5) measured by the acid hydrolysis method.The results on the fatty acid profile measured by gas chromatography indicate that the four most abundant fatty acids are found to be oleic acid (19.7%), linoleic acid (23.5%), palmitic acid (20.9%) and stearic acid (11%). These four major fatty acids represent around 80 % of the total fatty acids. Other fatty acids are present at ≤ 6% each.

Table 5. *Lipid composition of extracted fibres from Fomes fomentarius. The average data presented in g per 100g of dry mass±SD*

Fatty acids	Mean, g/100 g dm	±SD
Oleic acid	19.74	5.20
Linoleic acid	23.48	7.72
Palmitic acid	20.94	1.99
Stearic acid	10.99	0.06
Crude fat	1.90	0.88

Crude fat in extracted fibres have around 33% of the same value in raw material. Thus, like melanin-like substances, the waxes, lipids, resins and other ethanol extractives will be removed during the extraction process.

Nutritional Value

According to the published data [62], the calorific value of dietary fibres is very low and can be estimated to be around 2 kcal/g of product coming solely from fermentation in the

colon. A negligible amount of other cell components cannot significantly affect the nutritional value of the copolymer (Table 7).

Table7. *The nutritional profile of extracted fibres from Fomes fomentarius*

Parameter	kcal/100 g
Carbohydrates except glucans (%, m/m)	≤ 0.5
Lipids (%, m/m)	≤ 2
Proteins (%, m/m)	≤ 2
Fibres	76
Total calorific value	180

CONCLUSIONS

Fomes fomentarius fruiting body is assembled of tiny long fibrous cells arranged in stable and elastic cell walls. The cell walls composed mainly of glucan, chitin and melanin-like substances (Fig. 3). As minor compounds hemicellulose clasters of mannan, galactan, xylan, arabinan and rhamnan were detected as well as uronic and glucuronic acids.

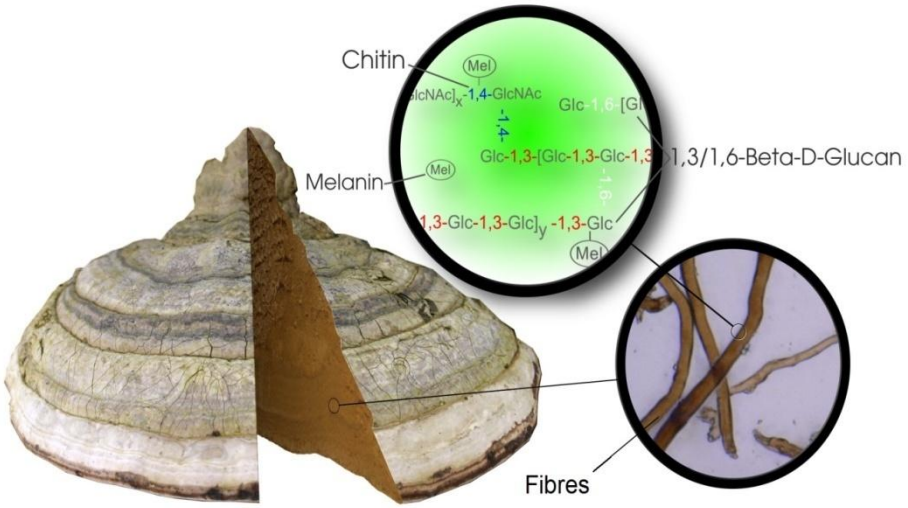

Figure 3. *Fomes fomentarius fruiting body with microscopic image of the isolated cell walls (fibres) and molecular structure of the main compounds of the cell wall (chitin, glucan and melanin).*

Mild purification of the milled fruiting bodies of the wood-decay fungus *Fomes fomentarius* leads to elimination of the bitter taste of the product and some modifications of the mushroom composition.

In comparison with raw material, the extracted fibres from *Fomes fomentarius* contain about one third of fats, a half of alpha-glucans, 25% less melanin-like substances, 15% less hemicellulose, around 25% more beta-glucans and 15% more chitin. High fibre content and respectively low calorific value were characteristics for the purified extracted fibres. Under severe extraction conditions a stronger modification of the cell walls leads to gradually increase of the chitin percentage in the cell walls.

Thus, after mild hot purification, the cell walls could be potentiated being more bio available and accessible for specific receptors, immune cells, intestinal microbiota etc. and could be a good candidate to replenish a number of vital mushrooms used as dietary supplements.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

[1] Wasser SP. Medicinal mushroom science: Current perspectives, advances, evidences, and challenges. Biomedical Journal.

2014;37(6):345-56. doi: 10.4103/2319-4170.138318.
[2] Lindequist U, Niedermeyer TH, Jülich WD. The pharmacological potential of mushrooms. Evidence Based Complementary and Alternative Medicine. 2005;2(3):285-99.
[3] Berg B, Lelley JI. Kompendium der Mykotherapie. Natura Viva Verlags GmbH,Germany; 2013.
[4] Ardigò W. Healing with Medicinal Mushrooms. A practical handbook. Youcanprint Self-Publishing, Italy; 2017.
[5] Hobbs C. Medicinal Mushrooms: An Exploration of Tradition, Healing, and Culture. Book Publishing Company; 2002.
[6] Roussel B, Rapior S, Charlot C, Masson CL, Boutié P. Histoire des utilizations therapeutiques de l'amadouvier, *Fomes fomentarius* (L. :Fr.) Fr. [History of the therapeutic uses of the tinder polypore, *Fomes fomentarius* (L.:Fr.)]. Revue d'histoire de la Pharmacie (Paris). 2002;50(336):599-614.
[7] Lelley JI. Die Heilkraft der Pilze: Wer Pilze isst lebt länger. BOSS Dr und Medien; 2008.
[8] Meuninck J. Basic Illustrated Edible and Medicinal Mushrooms. Rowman and Littlefield; 2015.
[9] Kim SH, Jakhar R, Kang SC. Apoptotic properties of polysaccharide isolated from fruiting bodies of medicinal mushroom *Fomes fomentarius* in human lung carcinoma cell line. Saudi Journal of Biolocigal Sciences 2015;22(4):484-90.
[10] Arpin N, Favre-Bonvin J, Steglich W. Le Fomentariol: Nouvelle Benzotropolone isolée de *Fomes fomentarius*. Phytochemistry. 1974;13:1949-1952.
[11] Seniuk OF, Gorovoj LF, Beketova GV, Savichuk NO, Rytik PG, Kucherov II, Prilutskaya AB, Prilutsky AI. Anti-Infective Properties of the Melanin-Glucan Complex obtained from *Fomes fomentarius*. International Journal of Medicinal Mushrooms. 2011;13(1):7-18.
[12] Gao HL, Lei LS, Yu CL, Zhu ZG, Chen NN, Wu SG. Immunomodulatory effects of *Fomes fomentarius* polysaccharides: an experimental study in mice [Article in Chinese] Nan Fang Yi Ke Da XueXueBao. 2009;29(3):458-461.
[13] Venckovsky BM, Tovstanovskaya VA, Bichkova NG, Priluckaya AB, Gorovoj LF. Infected Wound Treatment In an Obstetric Practice with Use of the Preparation Mycoton. International Journal of Medicinal Mushrooms. 2001;3:243.
[14] Lee JS. Effects of *Fomes fomentarius* supplementation on antioxidant enzyme activities, blood glucose, and lipid profile in streptozotocin-induced diabetic rats. Nutrition research 2005;25(2):187-195.
[15] Robles-Hernandez L, Gonzales-Franco AC, Crawford DL, Chun WWC. Review of environmental organopollutants degradation by white-rot basidiomycete mushrooms. Tecnociencia Chihuahua. 2008;2(1):32-39.
[16] Beketova GV, Savichuk NO, Savichuk AV, Senyuk OF, Gorovoj LF, Alexeenko NV, Senyuk CV. Efficiency of the Mushroom Preparation Mycoton in Treatment of Chronic Lesions of the Upper Parts of the Digestive Tube. International Journal of Medicinal Mushrooms. 2001;3:116.
[17] Park YM, Kim IT, Park HJ, Choi JW, Park KY, Lee JD, Nam BH, Kim DG, Lee JY and Lee KT. Anti-inflammatory and Anti-nociceptive Effects of the Methanol Extract of *Fomes fomentarius*. Food and Nutrition. 2004;27(10):1588-93.
[18] Stamets PE, Naeger NL, Evans JD, Han JO, Hopkins BK, Lopez D, Moershel HM, Nally R, Taylor AW, Carris LM, Sheppard WS. Extracts of Polypore Mushroom Mycelia Reduce Viruses in Honey Bees. Scientific Reports. 2018;8, Article number: 13936.
[19] Liu L, Zhou S-B, Zheng WF. Inhibition of Tumor Cells by Ethanol Extract of *Fomes fomentarius* [J]. Carcinogenesis, Teratogenesis & Mutagenesis. 2005;17(2):104-106.
[20] Chen W, Zhao Z, Li Y. Simultaneous increase of mycelial biomass and intracellular polysaccharide from *Fomes fomentarius* and its biological function of gastric cancer intervention. Carbohydrate Polymers. 2011;85(2):369–375.
[21] Zang Y, Xiong J, Zhai WZ, Cao L, Zhang SP, Tang Y, Wang J, Su JJ, Yang GX, Zhao Y, Fan H, Xia G, Wang CG, Hu JF. Fomentarols A-D, sterols from the polypore macrofungus *Fomes fomentarius*. Phytochemistry. 2013;92:137-45.
[22] Seniuk OF, Gorovoj LF. Health Protection and Restoration under Low-Level Irradiation Conditions Using a Mycoton Preparation. International Journal of Medicinal Mushrooms. 2001;3:219-220.
[23] Seniuk OF, Gorovoj LF, Kovalev VA, Palamar LA, Krul' NI, Zhidkov AV, Chemerskij GF, Kireev SI, Khatuntseva IV. Features of behavioral reactions of chronically irradiated mice in the raised crosswise labyrinth with various genetically determined radio sensitivity and possibilities of their modification by the fungal biopolymer complex. [Article in Russian] Radiats Biol Radioecol. 2013;53(2):170-82.
[24] Grienke U, Zöll M, Peintner U, Rollinger JM. European medicinal polypores-a modern view on traditional uses. Journal of Ethnopharmacology. 2014;154(3):564-83.
[25] Neifar M, Laouani A, Chaabouni SE. The potent pharmacological mushroom *Fomes fomentarius*. Cultivation processes and biotechnological uses. In: Gupta VK. (eds.). Applications of Microbial Engineering. CRC Press. 2013. P. 300-322.

[26] Patel S, Goyal A. Recent developments in mushrooms as anti-cancer therapeutics: a review. Biotechnology. 2012; 2:1-15.
[27] Gorovoj LF, Kosyakov VN. Mycoton – new chitin materials produced from fungi. In: Karnicki ZS. (eds.) Chitin World, Wirtschaftsverlag NW, Bremerhaven, 1995. p. 632-647.
[28] Gorovoj L, Burdyukova L. Chitin produced from fungi: medicine application perspectives. In: Domard A. (eds.) Advances in Chitin Science, 1. Jacques Andre Publisher, Lyon, 1996. p. 430-440.
[29] Gorovoj LF, Seniuk OF, Beketova GV, Savichuk NO, Amanbaeva G. Use of the chitin-containing preparation Mycoton in pediatric gastroenterology. In: Muzzarelli RAA. (eds.) Chitosan per os; from dietary supplement to drug carrier. Atec, Italy, 2000. p. 201-221.
[30] Gorovoj L, Seniuk O, Beketova G, Savichuk N, Tarasenko P, Savichuk A, Alexeenko N, Seniuk K, Bulgakova I. Treatment of Helicobacter, Herpes and Candida infections of the digestive tract. In: Muzzarelli RAA, Muzzarelli C. (eds.) Chitosan in pharmacy and chemistry. Atec, Italy, 2002. p. 151-155.
[31] Alvarado P, Moreno G, Vizzini A, Consiglio G, Manjón JL, Setti L. Atractosporocybe, Leucocybe and Rhizocybe: three new clitocyboid genera in the Tricholomatoid clade (Agaricales) with notes on Clitocybe and Lepista. Mycologia. 2015;107(1):123-136.
[32] Kalitukha L. The Tinder Fungus and the Secret of the GFP-Complex: 3x Daily to Be Healthy and Happy - Without Side Effects. On Demand Publishing, LLC-Create Space, 2017.
[33] ICC Standard Method 156: Determination of Total Dietary Fibre.
[34] Sluiter A, Ruiz R, Scarlata C, Sluiter J, Templeton D. Determination of Extractives in Biomass. National Renewable Energy Laboratory. U.S. Department of Energy. Technical Report NREL/TP-510-42619.
[35] Algermissen B, Nündel M, Riedel E. Analytik von Aminosäuren mit Fluoreszenz-HPLC. GIT Fachzeitschrift Lab. 1989;33:783-790.
[36] Kjeldahl J. Neue Methode zur Bestimmung des Stickstoffs in organischen Körpern (New method for the determination of nitrogen in organic substances). Zeitschrift für analytische Chemie. 1883;22:366-383.
[37] Bauer Petrovska B. Protein fraction in edible Macedonian mushrooms. European Food Research and Technology. 2001;212:469-472.
[38] Braaksma A, Schaap DJ. Protein analysis of the common mushroom *Agaricus bisporus*. Postharvest Biology and Technology. 1996;12:119-127.
[39] Sluiter A, Hames B, Ruiz R, Scarlata C, Sluiter J, Templeton D, Crocker D. Determination of structural carbohydrates and lignin in biomass. Laboratory analytical procedure (LAP). U.S. Department of Energy. Technical Report NREL/TP-510-42618. 2008, Revised August 2012.
[40] Ekblad A, Näshom T. Determination of chitin in fungi and mycorrhizal roots by an improved HPLC analysis of glucosamine. Carbohydrate Research. 1996;178(1):29–35.
[41] Chen GC, Johnson BR. Improved colorimetric determination of cell wall chitin in wood decay fungi. Applied Environenmental Microbiology. 1983;46(1):13-16.
[42] Megazyme International Ireland Ltd. Mushroom and yeast ß-glucan assay procedure booklet. K-YBGL 12/16. Wicklow (Ireland): Megazyme International Ireland Ltd.:2016.
[43] Müller E, Löffler W. Mykologie – Grundriss für Naturwissenschaftler und Mediziner. 5. Auflage, Georg Thieme Verlag, Stuttgart, New York 1992.
[44] Wakamatsu K, Ito S. Advanced chemical methods in melanin determination. Pigment Cell Research. 2002;15:174-183.
[45] Eisemann H, Casadevall A. Synthesis and assembly of fungal melanin. Applied Microbiology and Biotechnology. 2012;93:931-940.
[46] Scholz M, Leupold D. Ortsaufgelöstes Messverfahren für die Detektion von Melanin in Fluorophorgemischen in einer Festkörperprobe. Patentschrift DE10 2006 029 809.
[47] Jaehrig S, Rohn S, Kroh LW, Fleischer LG, Kurz T. In vitro potential antioxidants activity of (1-3),(1-6)-ß-D-glucans and protein fractions from *Saccharomyces cerevisiae* cell walls. Journal of Agriculture and Food Chemistry. 2007;55:4710-16.
[48] Bak WC, Park JH, Park YA, Ka KH. Determination of glucan in the fruiting bodies and mycelia of *Lentinula edodes* cultivars. Mycobiology. 2014;42:301-04.
[49] Chatterjee A, Khatua S, Chatterjee S, Mukherjee S, Mukherjee A, Paloi S, Acharya K, Bandyopadhyay SK. Polysaccharide-rich fraction of Termitomyceseurhizus accelerate healing of indomethacin induced gastric ulcer in mice. Glyconjunction Journal. 2013;30:759-68.
[50] Sari M, Prange A, Lelley J, Hambitzer R. Screening of beta-glucan contents in commercially cultivated and wild growing mushrooms. Food Chemistry. 2017;216:45-51.
[51] Bach F, Helm CV, Bellettini MB, Maciel GM, Haminiuk CWI. Edible mushrooms: a potential source of essential amino acids, glucans and minerals. Food Science and Technology. 2017;52:2382-92.
[52] Majtan J. Pleuran (ß-glucan from *Pleurotus ostreatus*): an effective nutritional supplement against upper respiratory tract infections? Medicine and Sport Science. 2012;59:57-61.

[53] Lee DH, Kim HW. Innate immunity induced by fungal β-glucans via dectin-1 signaling pathway. International Journal of Medicinal Mushrooms. 2014;16(1):1-16.
[54] Young SH, Dong WJ, Jacobs RR. Observation of a partially opened triple-helix conformation in 1-3-beta-glucan by fluorescence resonance energy transfer spectroscopy. Journal of Biological Chemistry, 2000;275(16):11874-11879.
[55] Tsuzuki A, Ohno N, Adchi Y, Yadomae T. Interleukin 8 production by human leukocytes stimulated by triple or single helical conformer of an antitumor (1-3)-beta-D-glucan preparation, Sonifilan. Drug Development Research, 1999;48:17-25.
[56] Ohno N, Hashimoto Y, Adachi Y, Yadomae T. Conformation dependency of nitric oxide synthesis of murine peritoneal macrophages by beta-glucans in vitro. Immunological Letters, 1996;52:1-7.
[57] Tyler HL, Haron MH, Pugh ND, Zhang J, Jackson CR, Pasco DS. Bacterial components are the major contributors to the macrophage stimulating activity exhibit by extracts of common edible mushrooms. Food and Function. 2016;7: 4213-4221.
[58] Nud`ga LA. Structural and chemical modification of chitin, chitosan and chitin-glucan complexes. 2006. Dissertation Habil. https://vivaldi.nlr.ru/bd000169484/view#page=16
[59] Osovskaya II, Budilina DL, Tarabukina EB, Nud`ga LA. Chitin-glucan complexes. Physical-chemical properties and molecular characteristics. Ed. Poltorazkii G.M. Textbook. Saint Petersburg. 2010.
[60] Shah H, Khalil IA, Jabeen S. Nutritional composition and protein quality of *Pleurotus mushroom*. Sarhad Journal of Agriculture. 1997;13:621-627.
[61] EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA); Scientific Opinion on the safety of "Chitin-Glucan" as a Novel Food ingredient. EFSA Journal 2010;8(7):1687.
[62] EFSA Panel on Dietetic Products, Nutrition and Allergies (NDA); Scientific Opinion on Dietary Reference Values for carbohydrates and dietary fibre. EFSA Journal 2010;8(3):1462.

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