BIOLOGICALLY ACTIVE SUBSTANCES FROM FRUITING BODIES AND MYCELIA OF MEDICINAL MUSHROOM *LENTINUS EDODES* (BERK.) SING.

V. G. BABITSKAYA[1];
N. A. BISKO[2];
N. YU. MITROPOLSKAYA[2]
[1]Institute of Microbiology, National Academy of Sciences of Byelorussia
[2]M.G.Kholodny Institute of Botany, National Academy of Sciences of Ukraine

Lentinus edodes (Berk.)Sing. has excellent nutritional value and medicinal qualities [6, 10]. The shiitake mushroom is used as dietary supplements in the form of tablets, extracts etc. [14]. The bioactive polysaccharides of *L.edodes* or polysaccharides-protein complexes is applicated as biological response modifiers for inhibiting tumor growth and other potent therapeutic uses [11].

The aim of our work was comparative analysis of the mycelia and fruiting bodies chemical composition of different shiitake strains.

Materials and methods

4 strains of *Lentinus edodes* (Berk.) Sing. were obtained from the culture collection of the Institute of Microbiology of the National Academy of Sciences of Byelorussia, Minsk and Institute of Botany of the National Academy of Ukraine, Kiev.

Mycelium of these strains was grown in submerged conditions on the glucose-peptone nutrient medium, (g/l): glucose - 10, peptone - 3, K_2HPO_4 - 1, KH_2PO_4 - 1, $MgSO_4 \bullet 7H_2O$ - 0.25, corn extract - 20 ml, deionized water - 1000 ml, pH 5.5.

After preparation the medium was sterilized by autoclaving for 20 min at 121°C. Mycelia were grown in 5-l flasks on the orbital shaker. Seed culture for 5 l flasks was developed in 0.5-l flasks containing 0.05l of cultivation medium inoculated with homogenized mycelia from Petri dishes. The biomass was ready to harverst after 7 days of cultivation at 28°C.

Mycelia of *L. edodes* strains were separated from medium by filtration. Mycelium was washed off with distilled water, dried to at 60°C, and pounded.

Fruiting bodies of *L. edodes* were grown on mixture of oak sawdust with wheat bran (4:1) [2]. Fruiting bodies of *L. edodes* were harvested, dried at 60°C and pounded.

The content of true protein in fruiting bodies and mycelia of *L. edodes* was estimated according to Lowry's method [9], chitin-glucan complex according to Kurshner and Ganek's method [12]. Lipids were extracted according to Folch's method [4]. The content of amino acids was estimated on amino acid analyzer AAA-881 "Microtechna" [8] fatty acids - on gas-liquid chromatograph "Chrom-5" with 15% polyethylenglycol succinate as liquid (temperature of column is 160°C, temperature of evaporation - 210°C) [7, 13[2]]. Exo- and endopolysaccharides were determined in the cultured liquid, mycelia and fruiting bodies of *L. edodes* [3, 5].

Results and discussion

Analysis of data given in Table 1 showed that the mycelia of the studied strains of *L. edodes* slightly differ one from other by main indexes of the chemical composition. However, the content of the true protein and lipids in the mycelia was higher than in fruiting bodies on 17-53% and on 95-96% accordingly for all strains. It should be noted that fruiting bodies of *L. edodes* contained greater quantity of chitin-glucan complex and phenolic substances.

Table 1

The chemical composition of *Lentinus edodes* strains

Strain	Bio-mass, g/l	Lipids, % a.d.m.	True protein, % a.d.m.	Complex of chitin-glucans, % a.d.m.	Polysaccharides		Phenolic substan-ces, mg %
					Endopoly-saccha-rides, % a.d.m.	Exopoly-saccha-rides, g/l	
101 m	5.7	7.5	20.0	7.6	3.2	3.5	1660
182 m	8.0	9.0	23.0	6.0	3.5	4.3	1800
182 f	-	0.3	11.0	11.0	3.2	-	2100
185 m	7.8	8.7	23.0	5.8	3.2	3.4	1630
185 f	-	0.5	19.1	10.2	3.0	-	1920
192 m	6.2	6.9	22.5	7.4	2.8	2.4	1500

m – mycelia
f – fruiting bodies

Earlier it has been demonstrated, that the content of phenolic substances in fruiting bodies of *Pleurotus ostreatus* was also higher but the content of lipids - lower in comparison with mycelia too [1].

It should be noted that the high differences between the mycelia and fruiting bodies of *L. edodes* strains were related with the content of the complex of the chitin-glucan substances – the fruiting bodies contented them almost 2 times higher compared to the mycelia.

The protein composition of the mycelia of studied strains included 17 amino acids (Table 2). The greatest content was characterized for aspartic and glutamic acids. Difference in content of some individual amino acids between *L. edodes* strains was considerable. For example, the content of lysine of strain 101 was on 65% greater than in strain 182, histidine - three times, methionine – two times, tyrosine – 4.4 times (Table 2). The fruiting bodies and mycelia of strain 185 differed on the content of some aminoacids: the fruiting bodies contained the greater quantities of lysine, histidine, argenine, threonine, methionine, tyrosine, and the mycelium – the higher quantities of aspartic acid, glycine, alanine, cystine, leucine, isoleucine (Table 2).

Table 2

The content of amino acids in proteins of *Lentinus edodes* strains (% of true protein)

Constituent	Strain 101, mycelia	Strain 182, mycelia	Strain 185		Strain 192, mycelia
			mycelia	fruiting bodies	
lysine	2.1	4.9	2.0	6.0	2.5
histidine	1.0	1.2	1.2	3.0	0.8
argenine	8.5	4.2	4.1	9.6	4.7
aspartic acid	10.7	10.0	9.9	7.0	11.5
threonine	6.3	4.5	4.8	7.2	5.8
serine	6.3	5.9	5.6	5.7	6.3
glutamic acid	17.6	17.0	22.9	20.0	18.3
proline	5.2	5.0	5.4	5.5	5.7
glycine	6.0	5.2	5.3	3.4	6.1
alanine	7.0	6.1	6.6	4.7	8.1
cystine	3.4	3.2	3.2	2.0	2.5
valine	6.5	6.9	6.4	6.9	6.5
methionine	1.2	2.2	1.2	2.4	1.2
isoleucine	4.3	5.2	5.0	3.3	4.2
leucine	9.3	10.0	8.4	5.1	7.5
tyrosine	1.4	4.5	4.0	6.2	4.0
phenylalanine	3.2	4.0	4.0	3.7	4.3

The data obtained indicated that the lipids of mycelia of all strains of *L. edodes* consist of 10 fatty acids (Table 3). The highest content was characterized for linoleic ($C_{16:0}$) and palmitic ($C_{18:2}$) acids. Unlike from the mycelium of *P. ostreatus*, grown on the same medium, the mycelia of *L. edodes* had in their composition myristic ($C_{14:0}$), palmitoleic ($C_{16:1}$) and heptadecenoic ($C_{17:1}$) acids [1]. It was found that palmitoleic ($C_{16:1}$) and heptadecenoic ($C_{17:1}$) were absent in the fruiting bodies of *L. edodes* as compared with the mycelium of the same strain (table 3). In contrast to *P. ostreatus*, mycelia of *L. edodes* have the higher content of unsaturated fatty acids than fruiting bodies [1].

Table 3

The content of fatty acids in the lipids of *Lentinus edodes* strains (% of total lipids)

	101 m	**182 m**	**185 m**	**185 f**	**192 m**
$C_{14:0}$	0.90	0.73	0.66	1.20	0.92
$C_{15:0}$	1.37	1.58	1.32	1.40	2.12
$C_{16:0}$	18.99	21.53	19.72	28.20	22.08
$C_{16:1}$	2.05	0.85	0.88	-	0.82
$C_{17:0}$	0.74	0.45	0.55	-	0.54
$C_{17:1}$	0.49	0.45	0.88	traces	0.74
$C_{18:0}$	3.27	2.30	2.74	7.30	4.00
$C_{18:1}$	8.98	3.79	7.07	6.30	5.66
$C_{18:2}$	61.12	67.42	66.18	54.30	62.47
$C_{18:3}$	2.08	0.90	traces	1.40	0.64
Saturated fatty acids	25.28	26.59	24.99	38.10	29.67
Unsaturated fatty acids	74.72	73.41	75.01	61.90	70.33
Ratio of unsaturated fatty acids to saturated fatty acids	1.40	1.43	1.41	1.60	1.34

m – mycelia; f – fruiting bodies

We analyzed the carbohydrate composition in polysaccharides of the mycelia and fruiting bodies of *L. edodes* strains. It has been demonstrated that glucose is the main carbohydrate component as in the mycelia as in the fruiting bodies (table 4). The arabinosa was absented in the mycelia and was discovered in the trace quantities in the fruiting bodies of the all strains, with the exception of strain 185 (Table 4).

Table 4

The content of carbohydrates in polysacchrides of *Lentinus edodes* strains, % of total

Carbo-hydrate	Strain 101			Strain 182		
	Mycelia		Fruiting bodies	Mycelia		Fruiting bodies
	Endopoly-saccharides	Exopoly-saccharides		Endopoly-saccharides	Exopoly-saccharides	
arabinose	-	-	traces	-	-	traces
xylose	1.02	1.36	traces	traces	traces	-
mannose	8.16	-	5.50	10.25	16.82	4.78
galactose	8.38	traces	6.80	16.42	traces	4.82
glucose	82.44	98.64	87.70	73.33	89.18	90.40
	Strain 185			Strain 192		
arabinose	-	-	2.40	-	-	traces
xylose	2.15	traces	traces	2.00	1.5	-
mannose	10.65	16.20	6.87	10.60	14.20	6.20
galactose	12.69	3.44	6.64	10.80	traces	5.60
glucose	74.51	80.36	84.09	76.60	84.30	88.20

" - " - is absent

At the same time xylosa was estimated in the polysaccharide composition of mycelia, and was absented or presented in trace quantities in fruiting bodies. Exopolysaccharides differed from endopolysaccharides in the main on the small or trace quantities of the galactose in the carbohydrate composition.

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Biologically active substances from fruiting bodies and mycelia of medicinal mushroom *Lentinus edodes* (Berk.) Sing.

Babitskaya V. G., Bisko N. A., Mitropolskaya N. Yu

True protein, phenols, exo- and endopolysaccharides, amino acid and fatty acid content and composition of fruiting bodies and mycelia of 4 strains of medicinal edible mushroom *Lentinus edodes* (Berk.) Sing. were investigated. It has been estimated that the content of the true protein and lipids in the mycelia was higher, than in fruiting bodies, but the fruiting bodies contained greater quantity of chitin-glucan complex and phenolic substances. It has been demonstrated that the content of unsaturated fatty acids in mycelia was higher than in fruiting bodies.