

BRIEF COMMUNICATIONS

ORGANIC ACIDS AND CARBOHYDRATES FROM *Laetiporus sulphureus* FRUITING BODIES

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UDC 547.915:615.43

In continuation of research on the chemical composition of *Laetiporus sulphureus* (Bull.:Fr.) Murr fruiting bodies [1], we studied organic acids and analyzed preliminarily the carbohydrate components.

We used a culture of *L. sulphureus* (LS-BG-0804 strain) isolated and grown as before [1]. Fruiting bodies were grown under laboratory conditions in chambers at 21°C without low-temperature stimulation on sterile sunflower shell substrate. Immature fruiting bodies (primordia) appeared 14 d after the substrate was fully colonized with regular air-spray moistening. Methods for organic acids [2]; free carbohydrates, water-soluble polysaccharides (WSPS), and base-soluble polysaccharides (BSPS) [3]; mannite [4]; and chitin [5] were used for quantitative analysis.

Chromatographic analysis [PC, BuOH:HCOOH (85%), 3:1, triple development to a height of 30 cm, detection by bromocresol green, bromophenol blue, and KMnO₄] of *L. sulphureus* extracts showed five organic acids, tartaric (Tar), citric (Cit), malic (Mal), malonic (Mln), and succinic (Suc). These were isolated [6] and identified by melting point, molecular weight, optical rotation, and IR spectroscopy. A study of the accumulation dynamics of the organic acids found that their total content was 1.21-5.05%, reaching a maximum in aged fruiting bodies (Table 1). The greatest concentration of free acids (1.44%) was observed in ripe fruiting bodies; of bound acids (3.80%), toward the end of the life cycle. The dominant acid during the whole development period was Tar (1.30-1.52%).

Carbohydrate components of ground fruiting bodies (100 g) were studied by defatting in a Soxhlet apparatus using CHCl₃:C₂H₅OH (2:1) and extracting the solid with ethanol (80%, 1:20, 100°C, ×3). Chromatography [PC, isopropanol (80%), detection by KMnO₄-NaIO₄-benzidine] found three compounds (trehalose, arabite, mannite). The last was isolated after recrystallization of the fraction from MeOH (7.65% yield of the fraction mass) and identified by melting point, molecular weight, optical rotation, and IR spectroscopy.

After the alcohol-soluble components were removed, the raw material was extracted with H₂O (1:20, 100°C, ×4). The extract was concentrated and precipitated with ethanol (95%, 1:5). The resulting precipitate of WSPS was separated by centrifugation and dried. Yield, 3.67% of fruiting body mass. The WSPS did not react with iodine, $[\alpha]_D^{20} -24^\circ$ (c 0.05, H₂O). The IR spectrum exhibited absorption bands for pyranose ring (810 cm⁻¹), α - (840) and β -glycosidic bonds (890), and protein (1590). Total hydrolysis of the WSPS (2 M TFA, 100°C, 6 h) produced fucose, mannose, glucose, and galactose (1.0:7.3:8.4:8.5) and traces of xylose, rhamnose, and arabinose (HPTLC-densitometry [7]).

The raw material remaining after extraction with H₂O was treated with KOH (5%, 1:12, 20°C, ×2). The combined extract was acidified with H₂SO₄ (50%) until the pH was 5-6. The resulting precipitate of BSPS was separated by centrifugation, washed with acetone, and dried. Yield, 41.46% of fruiting body mass. BSPS also did not react with iodine. Total hydrolysis produced only glucose. Solutions had negative optical rotation $[\alpha]_D^{20} -16^\circ$ (c 0.4, 5% KOH). The IR spectrum was similar to that of nonstarchy β -glucans from basidiomycetes [8].

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TABLE 1. Composition of Organic Acids from *L. sulphureus* Fruiting Bodies, %

Fruiting body phase	Parameter			Acid				
	X _f	X _b	X _Σ	Tar	Cit	Mal	MIn	Suc
Primordia	1.21	2.06	3.27	1.39	0.45	1.12	0.21	0.10
Ripe	1.44	1.67	3.11	1.30	0.56	0.58	0.32	0.35
Aged	1.25	3.80	5.05	1.52	1.48	1.32	0.28	0.45

X_f is the content of free acids; X_b, of bound acids; X_Σ, total content of organic acids.

TABLE 2. Composition of Carbohydrates from *L. sulphureus* Fruiting Bodies, %

Fruiting body phase	Mannite	Free carbohydrates	WSPS	BSPS	Chitin
Primordia	9.00	13.91	3.82	28.60	2.30
Ripe	14.94	15.87	5.46	25.62	2.00
Aged	25.21	27.18	2.81	33.53	4.90

The hydrolysis products of remaining tissue (conc. HCl, 100°C, 2 h) were glucosamine and traces of glucose (HPTLC, *i*-PrOH:CHCl₃:H₂O, 7:4:1, double elution, detection by ninhydrin). This indicated that *L. sulphureus* fruiting bodies contained chitin.

Studies of the composition of the carbohydrate complex in different development phases of *L. sulphureus* fruiting bodies showed that free carbohydrates (including mannite), BSPS, and chitin typically accumulated at the end of growth (Table 2). The WSPS content reached a maximum during ripening of fruiting bodies because of the need to store nutrients during basidiospore formation.

ACKNOWLEDGMENT

The work was performed with financial support from the Russian Foundation for Basic Research (Grant RFBR 08-04-98045).

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