

**INFLUENCE OF LOW-INTENSITY LIGHT
ON THE BIOSYNTHETIC ACTIVITY
OF THE MEDICINAL MACROMYCETE
Laricifomes officinalis (*Fomitopsidaceae*, *Polyporales*)
in vitro**

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Understanding the impact of artificial lighting on the biosynthetic and biological activity of medicinal mushrooms will help enhance technologies aimed at obtaining bioactive compounds.

Aim. The goal of our work was to determine the influence of low-intensity quasi-monochromatic and laser light on biosynthetic activity, including the antioxidant activity of the medicinal fungus *Laricifomes officinalis* under submerged cultivation conditions.

Methods. The effect of light on the biosynthetic activity of *L. officinalis* was studied using sources of low-intensity coherent monochromatic laser light and quasi-monochromatic radiation of light-emitting diodes (LEDs) with specified spectral-intensity characteristics.

Results. The most pronounced stimulating effect on the biosynthetic activity of the *L. officinalis* strain was observed when samples were irradiated with blue (488 nm laser and 470 nm LED) and red (650 nm LED) light. Under these conditions, there was an increase in the synthesis of mycelial mass, polysaccharides, and the quantity of total phenolic compounds. Low-intensity light irradiation caused changes in both the quantitative and qualitative composition of the fatty acid profile of the mycelial mass. Red light irradiation resulted in an increase in the quantity of polyunsaturated fatty acids. A correlation was established between the quantity of total phenolic compounds and antioxidant activity.

Conclusions. Obtained research results indicate low-intensity visible light as a promising regulator of the biosynthetic activity of *L. officinalis*, which can be useful in the biotechnology of its cultivation.

Key words: LED, laser, polysaccharides, fatty acids, total phenols, antioxidant activity, fatty acid.

Xylotrophic macrofungi represent a potentially promising group of fungal organisms for the development of mycobiototechnologies aimed at obtaining valuable bioactive metabolites and producing natural medicinal products based on them [1, 2]. Many species of xylotrophic macromycetes have long been recognized not only as functional food products but also as valuable sources for the development of nutraceuticals

and pharmaceuticals [3, 4]. The spectrum of revealed pharmacological activity exhibited by xylotrophic macromycetes is very broad. Depending on the expansion of knowledge about the chemistry, biotechnology, and molecular biology of fungi, as well as the improvement of screening methods (high-throughput screening, genomics, and proteomics), a rapid increase in the use of fungi for medical purposes can be expected.

One of the valuable species of xylotrophic macromycetes is *Laricifomes officinalis* (Vill.) Kotl. and Pouzar (syn. *Fomitopsis officinalis* (Vill.) Bondartsev & Singer), known as agarikon. The study of the mycochemical composition of not only the fruit bodies but also the mycelial mass of *L. officinalis* made it possible to isolate and identify various bioactive compounds, ranging from 60 to 115 according to different data sources [5–13]. Particularly valuable are secondary metabolites with confirmed pharmacological activity, including polysaccharides, lanostane-type triterpenoids, drimane-type sesquiterpenoids, sterols, coumarins, organic acids, phenolic compounds, indole compounds, flavonoids, and more [5, 7, 10]. The wide spectrum of pharmacological activity of *L. officinalis* has been confirmed both *in vivo* and *in vitro* [5, 8, 10, 14].

Considering the unique chemical composition and proven pharmacological activity of *L. officinalis*, intensification of submerged cultivation processes of this species using environmentally friendly factors, inter alia light, may have practical and scientific interest.

Experimental evidence has demonstrated that light and its parameters (wavelength, intensity, duration of exposure) significantly influence the adaptation processes of fungal organisms to the surrounding environment, including the growth of mycelium, its chemical composition, antioxidant, and antimicrobial activities [15–18]. The light-dependent nature of biosynthesis of certain compounds in fungi, such as carotenoids, melanins, nucleotides, amino, and fatty acids, has been confirmed [19, 20]. Studies on the impact of light influences on the vital functions of fungal organisms have been predominantly conducted on micromycetes [21, 22], while there is insufficient data on studying the specifics of the influence of quasi-monochromatic light from LED sources on the biosynthetic activity of medicinal macrofungi *in vitro*.

In our previous research, the influence of low-intensity laser irradiation on spore germination, growth processes, and fruit body formation of *L. officinalis* IBK 5004, *Lentinula edodes* (Berk.) Pegler, *Ganoderma lucidum* (Curtis) P. Karst., *Inonotus obliquus* (Fr.) Pilát was established [17, 18, 23–25]. Monochromatic low-intensity light based on light-emitting diodes (LEDs) and lasers serves as an environmentally friendly regulator of fungal organism vitality [26]. Understanding the physiology and assessing the ways to use LED and laser for influencing the metabolism

of *L. officinalis* can positively impact future research and provide knowledge for intensifying and regulating the cultivation processes of this valuable macrofungus species.

Intensification of the process of obtaining mycelial mass and polysaccharides under submerged cultivation conditions for *L. officinalis* has primarily been carried out through the optimization of nutrient medium composition [27]. However, photoregulation of biosynthetic activity for this species using low-intensity irradiation based on solid-state light-emitting diodes and lasers we carried out for the first time.

The aim of our study was to determine the impact of low-intensity quasi-monochromatic and laser light on the biosynthetic and antioxidant activity of the medicinal macromycete *L. officinalis* under submerged cultivation conditions.

Mushroom Strain. The subject of the research was a pure culture of the medicinal macromycete *L. officinalis* IBK 5004, preserved in the Mushroom Culture Collection at the M.G. Kholodny Institute of Botany of the National Academy of Sciences of Ukraine (acronym IBK) [28]. The taxonomic status of the *L. officinalis* IBK 5004 strain at the species level has been confirmed through molecular-genetic methods [29]. Data obtained are deposited in the NCBI database, available at GenBank (<https://www.ncbi.nlm.nih.gov/genbank/>), with accession numbers MF952886 for *L. officinalis* IBK 5004 isolates. Characteristic taxonomic features in the vegetative stage of the species were identified through previous cultural-morphological studies, allowing for the control of culture purity at all cultivation stages, and the composition of the liquid nutrient medium and cultivation conditions were established [27, 29].

The study on the influence of artificial low-intensity irradiation on the growth and biosynthetic activity of *L. officinalis* IBK 5004 was conducted following the scheme described by Mykchylova et al. [30].

The cultivation of the inoculum mycelium of *L. officinalis* IBK 5004 was performed on a liquid nutrient medium called Glucose-Peptone-Yeast (GPY), with the following composition per liter: glucose — 30.0 g; peptone — 3.5 g; yeast extract — 2.0 g; KH_2PO_4 — 1.0 g; K_2HPO_4 — 1.0 g; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ — 0.25 g; pH 5.5. Erlenmeyer flasks with a capacity of 0.5 l contained 100 ml of liquid GPY medium, shaken on a shaker at 120 rpm, at a temperature of 26 ± 1 °C in the dark for twelve days.

Impact of Quasi-Monochromatic Light. To assess the influence of quasi-monochromatic light as a factor affecting the biosynthetic activity of *L. officinalis* IBK 5004 mycelium, an artificial lighting system based on matrices of light-emitting diodes (LEDs) developed at the Institute of Physics of the National Academy of Sciences of Ukraine and an Argon Ion laser LGN-106M1 were used [24]. Light-emitting diodes with wavelengths $\lambda = 470$ nm (blue), $\lambda = 530$ nm (green), and $\lambda = 650$ nm (red) served as sources of quasi-monochromatic light. An argon ion laser LGN-106M1 emitting monochromatic radiation with a wavelength of 488 nm was used as a source of coherent visible light on the near wavelength. The experimental conditions were chosen to ensure equal energy doses of light exposure to the vegetative mycelium, with an energy density of 240 mJ/cm^2 at the sample surface in all experiments. The mycelium in the flasks was continuously irradiated. After irradiation, the mycelium was inoculated at a rate of 10% by volume into Erlenmeyer flasks containing 100 ml of liquid GPY medium for further cultivation under submerged culture conditions: shaken on a shaker at 120 rpm, at a temperature of 26 ± 1 °C.

After cultivation, the mycelial mass of *L. officinalis* IBK 5004 was separated from the culture liquid by filtration through Whatman No.1 filter paper. A weight of 2.0 grams of the mycelial mass was selected for determining the fatty acid profile. The remaining mycelium was dried to a constant weight in a forced-air oven (TCO-80 MICROMed, Shanghai Youding International Trade Co., LTD) at a temperature of 60.0 ± 0.1 °C. The quantity of dry biomass was calculated in grams per liter, taking into account the mass of the inoculum.

Determination of Intracellular Polysaccharides (IPS). Following the filtration of the culture liquid, the mycelium was washed with distilled water and dried to absolute dry biomass (a.d.b.) at 60 °C. For IPS extraction, the entire mycelium obtained from one flask was dried at 60 °C, homogenized in a laboratory blender, and then supplemented with distilled water (1:10 by weight). The mixture was boiled in a water bath (100 ± 1 °C) for 18 hours. Cytoplasmic contents were removed by multiple centrifugations (3,000 g for 15 minutes) of the homogenized mycelium suspended in distilled water. The washing procedure was stopped only when the optical density of the supernatant at 280 nm did not exceed 0.1 [31].

Determination of Extracellular Polysaccharides. The content of extracellular polysaccharides in the culture liquid was determined using the gravimetric method [31].

Determination of Fatty Acid Composition. The fatty acid composition of the mycelial mass samples was analyzed using the gas chromatography of methyl esters of fatty acids (ISO 5508:1990, IDT) with a gas chromatograph "Crystalux-4000M." A chromatographic column DB-225 (length = 30 m, diameter = 0.32 mm, and film thickness = 0.25 μm) with the sorbent (50% cyanopropylphenyl)-dimethylpolysiloxane (Agilent, USA) was utilized [32].

Preparation of Aqueous-Alcoholic Extracts of Mycelial Mass. Aqueous-alcoholic extracts (70% ethanol and methanol) were prepared with a ratio of 20 mg of dry mycelial mass to 1 ml of solvent. The mycelial mass was crushed, weighed, the solvent was added, and extraction was carried out on an ultrasonic bath at a temperature of 40 °C for 30 minutes. The extract was left in the refrigerator at 4 °C for 24 hours, after which the precipitate was filtered through Whatman No.1 filter paper. After filtration, the extract was centrifuged for 20 minutes at 13,500 g. The supernatant was used to determine the total amount of phenols and antioxidant activity. Each extract was prepared in triplicate.

Spectrophotometric Assays

Determination of Total Phenols Content. Total phenols of extracts were determined according to the method of Rašeta et al., [33]. The absorbance was measured at 750 nm using a spectrophotometer (U-1800, Hitachi Hightechnologies Co., Tokyo, Japan), with gallic acid used as a standard. The content of total phenols was calculated based on the calibration curve of gallic acid [the equation of standard curve: absorbance at 750 nm = $0.0025C_{\text{gallic acid}} (\mu\text{g/mL}) + 0.0982$, $R^2 = 0.985$]. Results were expressed as milligrams of gallic acid equivalents (GAEs) per gram of mushroom extract. The experiments were conducted in triplicate.

Determination of Radical scavenging activity (RSA)

The free radical scavenging activity of extracts *L. officinalis* in terms of the ability to annihilate 1-diphenyl-2-picryl-hydrazil (DPPH) was measured. For the RSA assay, the DPPH reagent (0.06 mM in ethanol) was added to the extract *L. officinalis*, and the mixture was kept in the dark for 30 min before the absorbance

was measured at 517 nm [34]. The higher RSA was indicated by lower absorbance. The RSA was calculated using the following equation:

$$\text{RSA (\%)} = (A_0 - A_1) / A_0 \times 100$$

where: A_0 — absorption of blank sample; A_1 — absorption of tested solution

Statistical analysis. Statistical data analysis was conducted using descriptive statistics, regression analysis, and analysis of variance (ANOVA) with the software Statistica 10.0. The significance of experimental data was assessed through ANOVA, considering results with p -values ≤ 0.05 as statistically significant. The experiments were conducted in triplicate. The quantitative content of fatty acids in the mycelial samples was determined by calculating the mean value across the three repetitions.

Results and Discussion

The impact of low-intensity quasi-monochromatic light on the synthesis of mycelial mass and polysaccharides. When determining the effectiveness of the effect of quasi-monochromatic light, we took into account the energy of the light quantum, light flux intensity, dose, and the spectral composition of light. LEDs and lasers were found to provide a spectral density that is unavailable with thermal light sources of comparable power.

In our study, it was observed that irradiation of the inoculated mycelium of *L. officinalis* IBK 5004 with low-intensity light during submerged cultivation led to an increase in the accumulation of mycelial mass and polysaccharides (Fig. 1). Irradiation with blue laser light ($\lambda = 488$ nm) and LED light ($\lambda = 470$ nm) allowed for the highest biomass yield, exceeding 14.7 g/L on the 12th day of cultivation, and showed no statistically significant differences. These results align with existing literature, where it has been experimentally established that blue light acts as a key signaling component, regulating gene expression and globally restructuring cellular metabolism in fungi [16, 35]. Previous studies from our research group demonstrated the influence of low-intensity laser light on the growth and fruiting of certain macrofungi species, including *Lentinula edodes*, *Pleurotus ostreatus* (Jacq.) P. Kumm., *Ganoderma lucidum* (Curtis) P. Karst [23, 25].

The accumulation of mycelial mass using red light ($\lambda = 650$ nm) reached 14.1 g/L, while irradiation with green light ($\lambda = 530$ nm) was the least effective, resulting in mycelial mass accumulation of 12.9 g/L.

Analysis of polysaccharide formation in all experimental variants revealed a correlation between the accumulation of mycelial mass and polysaccharides (Fig. 1). Irradiation of the inoculated mycelium with LEDs in all used wavelength ranges led to an increase in the synthesis of both exo- and endopolysaccharides. The most effective regimes were found to be irradiation with laser light at $\lambda = 488$ nm and LEDs at $\lambda = 470$ nm. Under these conditions, the accumulation of exopolysaccharides decreased by 47.6% and 42.8%, respectively. Irradiation with red ($\lambda = 650$ nm) and green ($\lambda = 530$ nm) light induced an increase in exopolysaccharide synthesis in *L. officinalis* IBK 5004 by 33.3% and 19.0%, respectively. The same trend was observed in the analysis of endopolysaccharide content in the mycelial mass.

Recently, polysaccharides from xylophagous macromycetes have begun to be used for various pharmacological and cosmetic applications [3, 36]. One of the most well-known examples is the anticancer drug based on the polysaccharide lentinan, obtained from *Lentinula edodes*. In Japan and China, lentinan is used as an adjuvant therapeutic agent in cancer treatment [37].

Our obtained results suggest that low-intensity light in the visible spectrum can be used in the biotechnology of submerged cultivation of *L. officinalis* IBK 5004 as a stimulator of biosynthetic activity, particularly in polysaccharide synthesis (Fig. 1). Due to their technical characteristics, LEDs and lasers have significant potential for application in the biotechnology of cultivating edible and medicinal macrofungi [26, 38].

Influence of low-intensity quasi-monochromatic light on the fatty acid profile of the mycelial mass. The next stage of our research aimed to determine the impact of the light factor on the fatty acid profile of the mycelial mass of *L. officinalis* IBK 5004. Comparative analysis of the lipid fraction of the mycelial mass revealed changes in the fatty acid profile of *L. officinalis* IBK 5004 under LED irradiation of different wavelengths.

In the investigated samples of mycelial mass, 21 fatty acids were identified, including nine saturated (SFA) and twelve mono- (MUFA) and polyunsaturated (PUFA) fatty acids (Table 1). The acids in the mycelium include those from the C_{14} – C_{24} series. MUFA and PUFA acids predominate, which is typical for basidiomycete macromycetes. The major fatty acids in all examined samples were linoleic acid ($C_{18:2}^{\omega_6}$) and oleic acid ($C_{18:1}$). Among saturated acids, palmitic acid

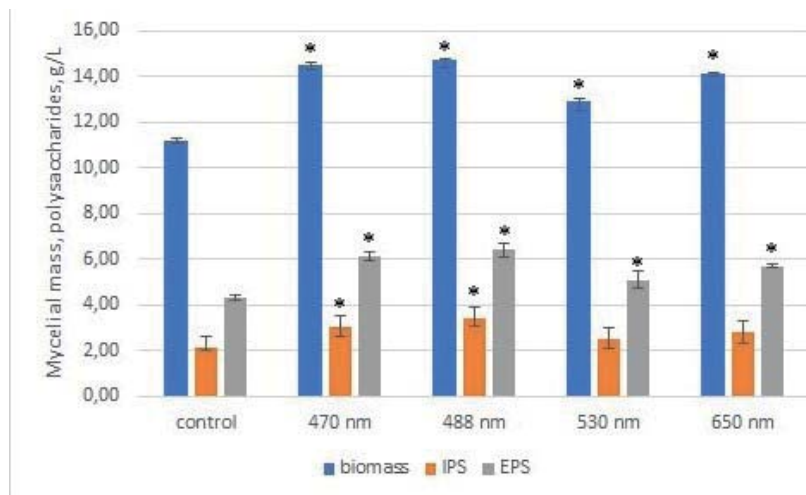


Fig. 1. The influence of low-intensity quasi-monochromatic light on the synthesis of mycelial mass and polysaccharides in *Laricifomes officinalis* IBK 5004

The data are presented as mean \pm standard error of the mean.

* $P < 0.05$ as compared to control (without irradiation)

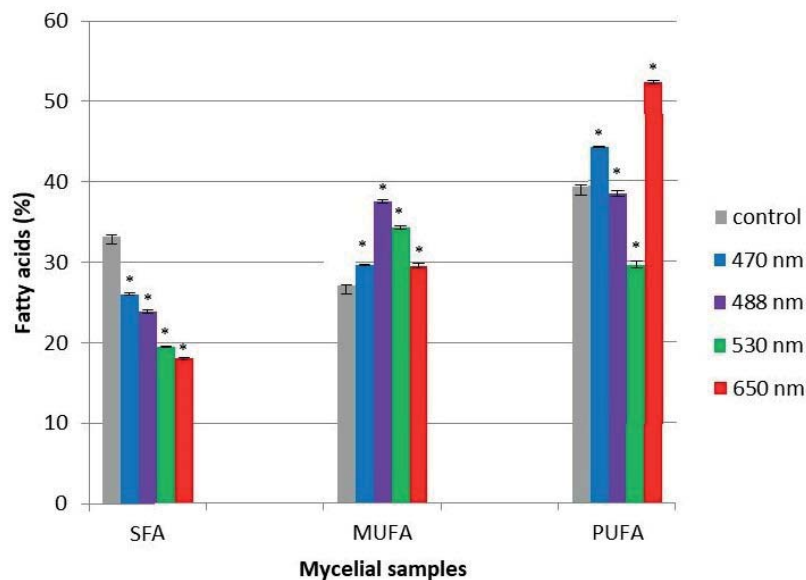


Fig. 2. The total content of saturated, monounsaturated, and polyunsaturated fatty acids in the mycelium of *L. officinalis* IBK 5004 under different irradiation modes

The data are presented as mean \pm standard error of the mean.

* $P < 0.05$ as compared to control (without irradiation).

($C_{16:0}$) and myristic acid ($C_{14:0}$) constitute the main portion. In the control sample (without irradiation), the SFA content was the highest, accounting for 33.25%. Under irradiation in all regimes, the SFA content decreased (Fig. 2). The lowest amount of SFA was recorded when irradiated with red light — 18.01%. Palmitic acid was the predominant component of SFA in all experimental variants, with its quantity varying from 14.01% under irradiation with $\lambda = 650$ nm to 24.81% in the control.

In the examined samples of *L. officinalis* IBK 5004 mycelium, three ω -9 MUFA ($C_{18:1 \omega-9}$, $C_{20:1 \omega-9}$, $C_{22:1 \omega-9}$), two ω -3 PUFAs ($C_{20:3 \omega-3}$, $C_{20:5 \omega-3}$), and three ω -6 PUFAs ($C_{18:2 \omega-6}$, $C_{20:2 \omega-6}$, $C_{20:4 \omega-6}$) were identified, belonging to the group of essential fatty acids. The synthesis of all ω -9 MUFA occurred when exposed to blue LED light. The greatest impact of LED irradiation on PUFA synthesis was recorded when exposed to red light ($\lambda = 650$ nm). The minimum content of linoleic acid (36.72%) was observed under laser light irradiation ($\lambda = 488$ nm). It is worth

noting that in the control sample (without irradiation), PUFA was represented by linoleic acid (C_{18:2 ω-6}) and a small amount of EPA (C_{20:5 ω-3}), while irradiation in all other used regimes contributed to the increase in newly formed fatty acids, improving the qualitative composition of PUFA. When analyzing data on the concentration of C₁₈ chain fatty acids,

oscillations in the levels of saturated (C_{18:0}) and unsaturated compounds (C_{18:1 ω-9}; C_{18:2 ω-6}) under different light regimes can be observed. All mentioned fluctuations in SFA/MUFA/PUFA content can be considered as a result of enzymatic reactions. In response to light exposure, fungal cells produce inducible enzymes, in this case, desaturases (catalyzing

Table 1

Fatty acid composition of the mycelial mass of *L. officinalis* IBK 5004 under different modes of irradiation with low-intensity quasi-monochromatic and laser light

No.	Fatty Acids	Irradiation modes				
		control	LED λ = 470 nm	LED λ = 530 nm	LED λ = 650 nm	Laser λ = 488 nm
Saturated Fatty Acids, content %						
1	Myristic acid (C _{14:0})	5.04	2.702	1.363	3.456	0.461
2	Pentadecanoic acid (C _{15:0})	ND	0.366	0.173	0.254	0.8154
3	Palmitic acid (C_{16:0})	24.81	20.81	15.28	14.01	21.35
4	Margaric acid (C _{17:0})	ND	0.096	0.062	0.087	0.0827
5	Stearic acid (C _{18:0})	0.765	0.521	0.188	0.221	0.987
6	Arachidic acid (C _{20:0})	1,982	0.717	0.326	ND	0.194
7	Behenic acid (C _{22:0})	ND	0.005	0.015	ND	ND
8	Трикозановая (C _{23:0})	0.628	0.438	ND	ND	ND
9	Lignoseriic acid (C _{24:0})	ND	0.599	0.359	ND	ND
	ΣSFA	33.25	26.05	19.46	18.01	23.89
Monounsaturated Fatty Acids (MUFAs), %						
10	Myristoleic acid (C _{14:1 ω-5})	1.061	0.792	0.376	ND	0.624
11	Pentadecenoic acid (C _{15:1})	ND	1.371	1.050	1.249	0.258
12	Palmitoleic acid (C _{16:1 ω-7})	ND	0.259	0.499	0.470	0.414
13	Heptadecenoic acid (C _{17:1})	0.447	0.891	0.839	0.754	0.623
14	Oleic acid (C_{18:1 ω-9})	25.21	26.15	31.29	27.10	35.60
15	Gondoic acid (C _{20:1 ω-9})	0.350	0.138	0.249	ND	ND
16	Erucic acid (C _{22:1 ω-9})	ND	0.033	ND	ND	ND
	ΣMUFA	27.06	29.64	34.30	29.57	37.52
Polyunsaturated Fatty Acids (PUFAs), %						
17	Linoleic acid (C_{18:2 ω-6})	38.55	43.88	44.91	51.96	37.47
18	Eicosadienoic acid (C _{20:2 ω-6})	ND	ND	0.017	0.017	ND
19	Arachidonic acid (C _{20:4 ω-6})	ND	0.012	0.042	0.152	0.955
20	EPA (<i>cis</i> -5,8,11,14,17-eicosapentaenoic acid, C _{20:5 ω-3})	0.862	0.321	0.273	0.235	ND
21	ETA (<i>cis</i> -8,11,14-eicosatrienoic acid, C _{20:3 ω-3})	ND	ND	0.714	0.004	ND
	ΣPUFA	39.41	44.22	45.96	52.37	38.42
	ΣMUFAs+ΣPUFAs	66.47	73.86	80.26	81.94	75.94
	ΣMUFAs+ΣPUFAs/ΣSFAs	1.99	2.8	4.1	4.5	3.17

Note. The average value among three repetitions was considered as the result of the fatty acid composition.

Table 2

Values of Total Phenolic Content (TPC) of the extracts mycelium *L. officinalis* IBK 5004
($n = 3, X \pm SD$)

Irradiation modes	TPC mg of GAE/g DM	
	water-ethanol extract	methanol extract
control	56.57±2.7	48.11±2.7
470 nm	81.23±3.4*	77.32±3.4*
488 nm	84.72±2.1*	80.73±2.1*
530 nm	66.34±3.2*	60.15±3.2*
650 nm	78.36±2.4*	70.21±2.4*

Note. The data are presented as mean \pm standard error of the mean.

* $P < 0.05$ as compared to control (without irradiation).

the conversion of single bonds between carbon atoms in acyl chains (C–C) to double bonds (C = C)). From the perspective of fungal metabolism, fatty acids are essential as precursors for aromatic compounds; for example, linoleic acid serves as a precursor to the alcohol compound 1-octen-3-ol, which imparts a specific aroma to fungi [39].

Thus, analyzing the obtained results, it can be concluded that irradiation with low-intensity quasi-monochromatic light affects the fatty acid profile of *L. officinalis* IBK 5004 mycelial mass. Changes occurred both in the quantitative and qualitative composition of components. The presence of a significant amount of PUFA, including essential ones such as oleic acid (C_{18:1} ω-9) and linoleic acid (C_{18:2} ω-6), in the composition of *L. officinalis* IBK 5004 mycelial mass classifies it as a valuable lipid source. Additionally, the presence of endopolysaccharides in the mycelial mass of *L. officinalis* IBK 5004, which have proven beneficial properties for the human body, may enhance the nutraceutical effect of this species.

The influence of low-intensity quasi-monochromatic light on the total content of phenolic compounds and antioxidant activity. The results of spectrophotometric studies showed a relatively high extraction yield and total phenolic content (TPC) in ethanol extracts of mycelial mass (Table 2). A wide range of TPC values was established, ranging from 48.11±2.7 to 84.72±2.1 mg of GAEs/g of dry mass, depending on the irradiation regimes (Table 2). The highest TPC values were found in ethanol extracts of *L. officinalis* irradiated with blue LED and laser light — 81.23±3.48 mg of GAEs/g of dry mass and 84.72±2.1 mg of GAEs/g of dry mass, respectively. The lowest values were recorded for irradiation with green light.

Recently, xylophilic macrofungi have been considered as a natural source of phenolic compounds with high antioxidant activity [33, 40]. The biological activity of phenolic compounds may be associated with their ability to chelate metals, inhibit lipoxygenase, and eliminate free radicals [41]. Many degenerative diseases are linked to the negative impact of free radicals, as they induce oxidative damage to DNA, proteins, and other macromolecules. These damages accumulate with age and are considered a primary form of endogenous damage to the body, leading to aging [33, 41]. Free radicals are neutralized by cellular defense systems, including phenolic compounds, which protect cells from oxidative damage. However, this is insufficient to completely prevent oxidative stress-induced damage. Therefore, exogenous dietary antioxidants or natural products based on medicinal mushrooms are promising for use as nutraceuticals in chronic diseases [2, 42].

Various phenolic compounds have been identified and quantified in the fruit bodies and mycelium of *L. officinalis* [5, 6, 43]. Additionally, a correlation has been established between antioxidant activity and the content of phenolic compounds in extracts of the fruit bodies of this species [10]. Similar correlations between Total Phenolic Content (TPC) and antioxidant activity have been found in many edible and medicinal macrofungi [40, 45].

The results of our study on mycelial extracts showed high Radical Scavenging Activity (RSA) against DPPH. The highest RSA values (96.06%) were determined for mycelial samples irradiated with blue light. The lowest values from all spectrophotometric analyses were observed for mycelium irradiated with green light (Fig. 3). Overall, the correlation between high RSA and the amount of Total Phenolic

Content was evident in all *L. officinalis* extracts. However, other antioxidants present in the investigated extracts may also contribute to RSA.

Our results regarding the spectral sensitivity of *L. officinalis* concerning Total Phenolic Content (TPC) and antioxidant activity align with findings from other researchers. Specifically, according to Jang et al., [26], the use of blue light ($\lambda = 475$ nm) in the cultivation of *Hypsizygus marmoreus* (Peck) H.E. Bigelow led to an increase in the yield of fruit bodies with high ergosterol content, polyphenols, and high antioxidant activity [26]. Additionally, Huang et al. [38] reported that when using LED blue light in the cultivation of *Lentinus sajor-caju* (Fr.) Fr., not only did the yield and nutritional value of fruit bodies increase, but the antioxidant activity of extracts from fruit bodies also saw an enhancement.

Damaso et al. [45] investigated the impact of LED on the growth, fruiting body formation, total phenolic content, and antioxidant activity of *Lentinus tigrinus* (Bull.) Fr. The highest phenolic content (25.04 mg GAEs/g sample) and radical scavenging activity (61.29%) were observed in the fruit bodies grown under blue and red LED irradiation.

Tiniola et al. [46] assessed the influence of LED on biomass accumulation, fruit body production, total phenolic content, and antioxidant properties of *Lentinus swartzii* Berk. The highest amount of mycelial mass was observed when the mycelium was irradiated with red light under submerged cultivation

conditions. The highest values of radical scavenging activities (73.95%) and TPC (26.08 mg GAEs/g sample) were found in the ethanol extract of the fruit body irradiated with red light-emitting diodes.

Thus, the results of the study on the photoresponses of the *L. officinalis* IBK 5004 strain suggest the possibility of implementing effective microbiotechnologies using LED and laser of specific wavelength. It has been established that short-term exposure of the mycelium to low-intensity light with energy density up to 230 mJ/cm² in the blue and red wavelength ranges increases biosynthetic activity, specifically the synthesis of mycelial mass, polysaccharides, phenolic compounds, and influences the fatty acid profile and antioxidant activity of the *L. officinalis* IBK 5004 strain. This may have prospects for targeted regulation of the biosynthetic activity of the strain.

Conclusion

It has been established that short-term exposure of mycelium to low-intensity light with an energy density of up to 230 mJ/cm² in the blue and red wavelength ranges increases the biosynthetic activity of the studied producer strain *L. officinalis* IBK 5004.

Selected modes of photostimulation of biosynthetic activity can be used in the biotechnology of submerged cultivation of the edible medicinal mushroom *L. officinalis* IBK 5004 to intensify the technological stages and

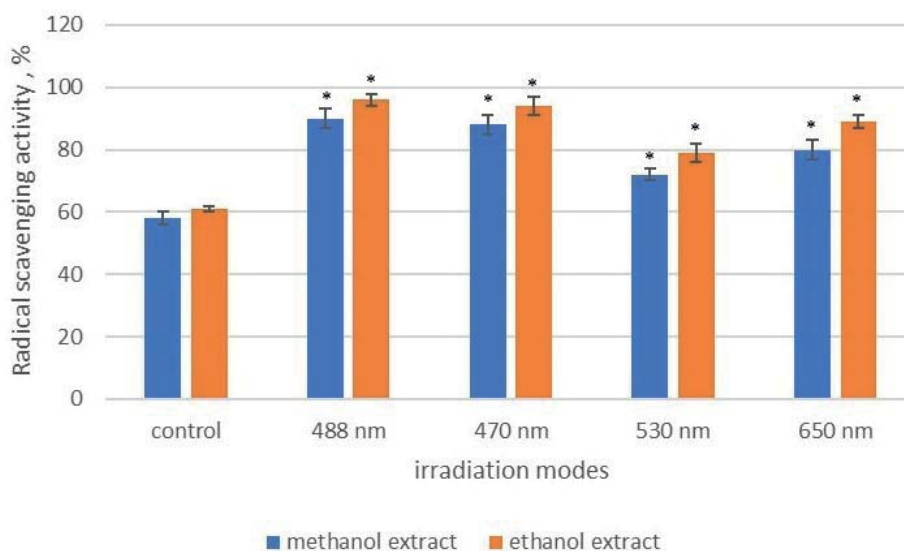


Fig. 3. Radical scavenging activity (RSA) of mycelial mass extracts of the *L. officinalis* IBK 5004
The data are presented as mean \pm standard error of the mean. * $P < 0.05$ as compared to control (without irradiation)

increase the yield of the final product (mycelial mass, polysaccharides, phenolic substances, mono- and polyunsaturated fatty acids).

The study of photobiological reactions of fungi and the accumulation of experimental material on the stimulating effect of low-intensity quasi-monochromatic light on the biosynthetic activity of *L. officinalis* IBK 5004 can bring us closer to understanding the fundamental mechanisms of the action of light on the mushroom.

Author Contributions

Author Contribution O. M. & N. P. planned the work, contributed to the article's conception, manuscript article writing, editing, and conducted a study under submerged cultivation, the effect of low-intensity light on mycelial mass fatty acid profile. A. N. defined the light sources design and its modes of operations, calculated the doses of irradiation

inoculum, carried out its irradiation, discussed the irradiation effects, and participated in the paper preparation. O.B. & Ya. P. conducted a study of polysaccharides' accumulation and performed a statistical analysis. N.S. provided the new literature data for review. All authors contributed to the manuscript's revision and read and approved the submitted version.

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REFERENCES

- Hyde K.D., Xu J., Rapior S., Jeewon R., Lumyong S., Niego A.G.T., Abeywickrama P.D., Aluthmuhandiram J.V.S., Brahamanage R.S., Brooks S., Stadler, M. The amazing potential of fungi: 50 ways we can exploit fungi industrially. *Fungal Diversity*. 2019, v.97(1):1–136. <https://doi.org/10.1007/S13225-019-00430-9>
- Niego A.G., Rapior S., Thongklang N., Raspé O., Jaidee W., Lumyong S., & Hyde K.D. Macrofungi as a Nutraceutical Source: Promising Bioactive Compounds and Market Value. *Journal of Fungi*. 2021, 7(5):397. <https://doi.org/10.3390/JOF7050397>
- Badalyan S.M., Borhani A. Medicinal, nutritional, and cosmetic values of macrofungi distributed in mazandaran province of northern iran (Review). *International Journal of Medicinal Mushrooms*. 2019, 21(11):1099–1106. <https://doi.org/10.1615/IntJMedMushrooms.2019032743>
- Deshmukh S.K., Sridhar K.R., & Badalyan S.M. Fungal Biotechnology Prospects and Avenues. 2022; CRC Press: 450. <https://doi.org/10.1201/9781003248316>
- Fijałkowska A., Muszyńska B., Sulowska-Ziaja K., Kała K., Pawlik A., Stefaniuk D., Matuszewska A., Piska K., Pękala E., Kaczmarczyk P., Piętka J., & Jaszek M. Medicinal potential of mycelium and fruiting bodies of an arboreal mushroom *Fomitopsis officinalis* in therapy of lifestyle diseases. *Scientific Reports*. 2020, 10(1). <https://doi.org/10.1038/s41598-020-76899-1>
- Muszyńska B., Fijałkowska A., Sulowska-Ziaja K., Włodarczyk A., Kaczmarczyk P., Nogaj E., & Piętka J. *Fomitopsis officinalis*: a Species of Arboreal Mushroom with Promising Biological and Medicinal Properties. *Chemistry and Biodiversity*. 2020, 17(6). <https://doi.org/10.1002/cbdv.202000213>
- Han J., Li L., Zhong J., Tohtaton Z., Ren Q., Han L., Huang X., & Yuan T. Officimalonic acids A-H, lanostane triterpenes from the fruiting bodies of *Fomes officinalis*. *Phytochemistry*. 2016, 130:193–200. <https://doi.org/10.1016/J.PHYTOCHEM.2016.05.004>
- Han J., Liu W., Li M., Gu Y., Zhang Y., & Yuan T. Lanostane triterpenoids from the fruiting bodies of *Fomes officinalis* and their anti-inflammatory activities. *Molecules*, 2020, 25(20). <https://doi.org/10.3390/molecules25204807>
- Gafforov Y., Muszyńska B., Sulowska-Ziaja K., Tomšovský M., Yarasheva M., Pecoraro L., Mykchaylova O., & Rapior S. *Laricifomes officinalis* (Vill.) Kotl. & Pouzar — FOMITOPSIDACEAE. 2023:1237–1251. https://doi.org/10.1007/978-3-031-23031-8_116
- Flores G.A., Cusumano G., Ianni F., Blasi F., Angelini P., Cossignani L., Pellegrino R.M., Emiliani C., Venanzoni R., Zengin G., Acquaviva A., Di Simone S.C., Libero M.L., Nilofar Orlando G., Menghini L., & Ferrante C. *Fomitopsis officinalis*: Spatial (Pileus and Hymenophore) Metabolomic Variations Affect Functional Components and Biological Activities. *Antibiotics*. 2023, 12(4). <https://doi.org/10.3390/ANTIBIOTICS12040766>
- Zhang H., Aisa H.A., Liu Y., Tohtahon Z., Xin X., & Abdulla R. Characterization and

- identification of chemical constituents in aqueous extract of *Fomes officinalis* Ames based on ultrahigh-performance liquid chromatography-tandem quadrupole-Orbitrap high-resolution mass spectrometry. *Phytochemical Analysis*. 2023. <https://doi.org/10.1002/pca.3295>
12. Vedenicheva N.P., Al-Maali G.A., Bisko N.A., Shcherbatiuk M.M., Lomborg M. L., Mytropolska N.Y., Mykchaylova O.B., & Kosakivska I.V. Comparative analysis of cytokinins in mycelial biomass of medicinal mushrooms. *International Journal of Medicinal Mushrooms*. 2018, 20(9):837–847. <https://doi.org/10.1007/s11142-013-9235-x>
 13. Vedenicheva N.P., Al-Maali G.A., Mytropolska N.Yu., Mykchaylova O.B., Bisko, N.A., Kosakivska I.V. Endogenous cytokinins in medicinal basidiomycetes mycelial biomass. *Biotechnologia Acta*. 2016, 9(1):55–63. <https://doi.org/10.15407/biotech9.01.055>
 14. Girometta C. Antimicrobial properties of *Fomitopsis officinalis* in the light of its bioactive metabolites: a review. *Mycology*. 2018, 10(1):32–39. <https://doi.org/10.1080/21501203.2018.1536680>
 15. Corrochano L.M. Fungal photoreceptors: Sensory molecules for fungal development and behaviour. *Photochemical and Photobiological Sciences*. 2007, 6(7):725–736. <https://doi.org/10.1039/b702155k>
 16. Corrochano L.M. Light in the Fungal World: From Photoreception to Gene Transcription and Beyond. *Annual Review of Genetics*. 2019, 53:149–170. <https://doi.org/10.1146/annurev-genet-120417-031415>
 17. Poyedinok N.L., Mykchaylova O., Negriyko, A., Dudka I.A., Vasilyeva B.F., Efremenkova O.V. Induction of antimicrobial activity of some macromycetes by low-intensity light. *Biotechnologia Acta*. 2015, 8(1):63–70. <https://doi.org/10.15407/BIOTECH8.01.063>
 18. Poyedinok N.L., Tugay T.I., Tugay A.V., Mykchaylova O.B., Negriyko, A. Influence of nitrogen concentration on photoinduced growth, enzymatic activity and melanine synthesis by *Inonotus obliquus* (Ach.:Pers.) Pilát. *Biotechnologia Acta*. 2019, 12(4):34–41. <https://doi.org/10.15407/biotech12.04.034>
 19. Tisch D., & Schmoll M. Light regulation of metabolic pathways in fungi. *Applied Microbiology and Biotechnology*, 2010, 85(5):1259–1277. <https://doi.org/10.1007/S00253-009-2320-1>
 20. Halabura M.I.W., Avelino K.V., Araújo N.L., Kassem A.S.S., Seixas F.A.V., Barros L., Fernandes Á., Liberal Á., Ivanov M., Soković M., Linde G.A., Colauto N.B., & do Valle J.S. Light conditions affect the growth, chemical composition, antioxidant and antimicrobial activities of the white-rot fungus *Lentinus crinitus* mycelial biomass. *Photochemical & Photobiological Sciences*, 2023, 22(3):669–686. <https://doi.org/10.1007/S43630-022-00344-7>
 21. Castrillo M., García-Martínez J., & Avalos J. Light-dependent functions of the *Fusarium fujikuroi* CryD DASH cryptochrome in development and secondary metabolism. *Applied and Environmental Microbiology*, 2013, 79(8):2777–2788. <https://doi.org/10.1128/AEM.03110-12>
 22. Avalos J., & Carmen Limón M. Biological roles of fungal carotenoids. *Current Genetics*. 2015, 61(3):309–324. <https://doi.org/10.1007/S00294-014-0454-X>
 23. Poyedinok N., Buchalo A., Negriyko A., Potemkina J., & Mykchaylova O. The Action of Argon and Helium-Neon Laser Radiation on Growth and Fructification of Culinary-Medicinal Mushrooms *Pleurotus ostreatus* (Jacq.:Fr.) Kumm., *Lentinus edodes* (Berk.) Singer, and *Hericium erinaceus* (Bull.:Fr.) Pers. *International Journal of Medicinal Mushrooms*. 2003, 5(3):8. <https://doi.org/10.1615/InterJMedicMush.v5.i3.70>
 24. Poyedinok N.L., Negriyko A., Bisko N.A., Mykchaylova O. Energy Efficient Systems of Artificial Lighting in Technologies of Edible and Medicinal Mushroom Cultivation. *Nauka ta Innovacii*, 2013, 9(3):46–56. <https://doi.org/10.15407/SCIN9.03.046>
 25. Poyedinok N.L. Use of artificial light in mushroom cultivation. *Biotechnologia Acta*, 2013, 6(6):58–70. <https://doi.org/10.15407/biotech6.06.058>
 26. Jang M.J., Lee Y.H., Ju Y.C., Kim S.M., & Koo H.M. Effect of Color of Light Emitting Diode on Development of Fruit Body in *Hypsizygus marmoreus*. *Mycobiology*. 2018, 41(1):63–66. <https://doi.org/10.5941/MYCO.2013.41.1.63>
 27. Mykchaylova O.B., Poyedinok N.L., & Shchetinin V.M. Screening of strains of the medicinal mushroom *Fomitopsis officinalis* (Vill.) Bondartsev & Singer promising for biotechnological use. *Innovative Biosystems and Bioengineering*. 2022, 6(3–4):110–118. <https://doi.org/10.20535/IBB.2022.6.3-4.271383>
 28. Bisko N., Lomborg M., & Mykchaylova O. *The IBK Mushroom Culture Collection of the M.G. Kholodny Institute of Botany*. IBK Mushroom Culture Collection. (2023). Version 1.5.
 29. Mykchaylova O.B., Bisko N.A., Sukhomlyn M.M., Lomborg M.L., Pasaylyuk M.V., Petrichuk Yu.V., Gryganskyi A.Ph. Biological peculiarities of a rare medicinal mushroom *Fomitopsis officinalis* (Fomitopsidaceae, Polyporales) on agar media and plant substrates. *Regulatory Mechanisms in Biosystems*. 2017, 8(4):469–475. <https://doi.org/10.15421/021772>

30. Mykchaylova O., Dubova H., Lomberg M., Negriyko A., Poyedinok N. Influence of low-intensity light on the biosynthetic activity of the edible medicinal mushroom *Hericium erinaceus* (Bull.: Fr.) Pers. *in vitro*. *Archives of Biological Sciences*. 2023, 75(4):40. <https://doi.org/10.2298/ABS230821040M>
31. Bisko N., Mustafin K., Al-Maali G., Suleimenova Z., Lomberg M., Narmuratova Z., Mykchaylova O., Mytropolska N., & Zhakipbekova A. Effects of cultivation parameters on intracellular polysaccharide production in submerged culture of the edible medicinal mushroom *Lentinula edodes*. *Czech Mycology*. 2020, 72(1):1–17. <https://doi.org/10.33585/CMY.72101>
32. Dubova H., Dotsenko N., Mykchaylova O., & Poyedinok N. Study of aromatic components in the course of initiating enzymatic reactions in the edible mushroom *Pleurotus ostreatus*. *Food Science and Technology*, 2022, 15(4). <https://doi.org/10.15673/FST.V15I4.2254>
33. Rašeta M., Popović M., Knežević P., Šibul F., Kaišarević S., & Karaman M. Bioactive Phenolic Compounds of Two Medicinal Mushroom Species *Trametes versicolor* and *Stereum subtomentosum* as Antioxidant and Antiproliferative Agents. *Chemistry & Biodiversity*. 2020, 17(12):e2000683. <https://doi.org/10.1002/CBDV.202000683>
34. Jaszek M., Osińska-Jarozuk M., Janusz G., Matuszewska A., Stefaniuk D., Sulej J., Polak J., Ruminowicz M., Grzywnowicz K., Jarosz-Wilkolazka A. New bioactive fungal molecules with high antioxidant and antimicrobial capacity isolated from *Cerrena unicolor* idiophasic cultures. *Biomed Res Int*. 2013, :497492. <https://doi.org/10.1155/2013/497492>.
35. Yu Z., & Fischer R. Light sensing and responses in fungi. *Nature Reviews Microbiology*. 2019; 17(1):25–36. <https://doi.org/10.1038/S41579-018-0109-X>
36. Badalyan S., & Rapior S. Perspectives of Biomedical Application of Macrofungi. *Current Trends in Biomedical Engineering & Biosciences*. 2020, 19(5). <https://doi.org/10.19080/CTBEB.2020.19.556024>
37. Zhang M., Zhang Y., Zhang L., & Tian Q. Mushroom polysaccharide lentinan for treating different types of cancers: A review of 12 years clinical studies in China. *Progress in Molecular Biology and Translational Science*, 2019, 163:297–328. <https://doi.org/10.1016/BS.PMBTS.2019.02.013>
38. Huang M.Y., Lin K.H., Lu C.C., Chen L.R., Hsiung T.C., & Chang W.T. The intensity of blue light-emitting diodes influences the antioxidant properties and sugar content of oyster mushrooms (*Lentinus sajor-caju*). *Scientia Horticulturae*, 2017, 218:8–13. <https://doi.org/10.1016/J.Scienta.2017.02.014>
39. Ribeiro B., Guedes de Pinho P., Andrade P. B., Baptista P., & Valentão P. Fatty acid composition of wild edible mushrooms species: A comparative study. *Microchemical Journal*, 2009, 93(1):29–35. <https://doi.org/10.1016/j.microc.2009.04.005>
40. Kozarski M., Klaus A., Jakovljevic D., Todorovic N., Vunduk J., Petrović P., Niksic M., Vrvic M.M., & Van Griensven L. Antioxidants of edible mushrooms. *Molecules*. 2015, 20(10):19489–19525. <https://doi.org/10.3390/molecules201019489>
41. Cheung L.M., Cheung P.C.K., & Ooi V.E.C. Antioxidant activity and total phenolics of edible mushroom extracts. *Food Chemistry*. 2003, 81(2):249–255. [https://doi.org/10.1016/S0308-8146\(02\)00419-3](https://doi.org/10.1016/S0308-8146(02)00419-3)
42. Reis F.S., Barros L., Martins A., & Ferreira I.C.F.R. Chemical composition and nutritional value of the most widely appreciated cultivated mushrooms: An interspecies comparative study. *Food and Chemical Toxicology*, 2012, 50(2):191–197. <https://doi.org/10.1016/J.FCT.2011.10.056>
43. Sulikowska-Ziaja K., Fijałkowska A., & Muszyńska B. Selected Species of Medicinal/Arboreal Mushrooms as a Source of Substances with Antioxidant Properties. In *Reference Series in Phytochemistry* 2022:95–121. Springer Science and Business Media B.V. https://doi.org/10.1007/978-3-030-78160-6_38
44. Dubost N., Ou B., & Beelman R. Quantification of polyphenols and ergothioneine in cultivated mushrooms and correlation to total antioxidant capacity. *Food Chemistry*, 2007. 105(2):727–735. <https://doi.org/10.1016/j.foodchem.2007.01.030>
45. Damaso E.J., Dulay R.M., Kalaw S., & Reyes R. Effects of Color Light Emitting Diode (LED) on the Mycelial Growth, Fruiting Body Production, and Antioxidant Activity of *Lentinus tigrinus*. *CLSU International Journal of Science & Technology*. 2018. <https://doi.org/10.22137/ijst.2018.v3n2.02>
46. Tiniola R.C., Pambid R.C., Bautista A.S., & Milton Dulay R.R. Light emitting diode enhances the biomass yield and antioxidant activity of Philippine wild mushroom *Lentinus swartzii*. *Asian J. Agric. Biol*, 2021, (2):202008435. <https://doi.org/10.35495/ajab.2020.08.435>

**ВПЛИВ СВІТЛА НИЗЬКОЇ ІНТЕНСИВНОСТІ НА БІОСИНТЕТИЧНУ АКТИВНІСТЬ
ЛІКАРСЬКОГО МАКРОМІЦЕТА *Laricifomes officinalis* (Fomitopsidaceae, Polyporales)
in vitro**

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Розуміння механізмів впливу штучного освітлення на біосинтетичну та біологічну активність лікарських грибів допоможе інтенсифікувати технології, спрямовані на отримання біоактивних сполук.

Мета. Наша робота була спрямована на визначення впливу LED і лазерного світла низької інтенсивності на біосинтетичну та антиоксидантну активності лікарського гриба *Laricifomes officinalis* за умов глибинного культивування.

Методи дослідження. Вплив світла на біосинтетичну активність *L. officinalis* досліджували за допомогою джерел низькоінтенсивного когерентного монохроматичного лазерного світла та квазімонохроматичного випромінювання світлодіодів — LED із заданими спектрально-інтенсивними характеристиками.

Результати. Найбільш виражену стимулювальну дію на біосинтетичну активність штаму *L. officinalis* спостерігали за опромінення зразків синім (488 нм лазер і 470 нм LED) та червоним (650 нм LED) світлом. За цих режимів відмічено підсилення синтезу міцеліальної маси, полісахаридів та кількості загальних фенольних сполук. Опромінення світлом низької інтенсивності спричиняло зміни як кількісного, так і якісного складу жирнокислотного профілю міцеліальної маси. Під час опромінення червоним світлом відбулося збільшення кількості поліненасичених жирних кислот. Встановлено кореляцію між кількістю загальних фенольних сполук і антиоксидантною активністю.

Висновки. Результати досліджень дають підстави вважати світло низької інтенсивності у видимій частині спектру перспективним регулятором біосинтетичної активності *L. officinalis* у біотехнології його культивування.

Ключові слова: LED, лазер, полісахариди, жирні кислоти, загальні феноли, антиоксидантна активність.