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EXPERIMENTAL ARTICLES

The Role of Exogenous Lipids in Lycopene Synthesis in the Mucoraceous Fungus *Blakeslea trispora*

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Abstract—The addition of plant oils to the growth medium stimulated growth and lipid synthesis in the fungus *Blakeslea trispora*. However, only oils with high content of linoleic and especially linolenic acid enhanced lycopene formation. The increase in lycopene formation was accompanied by accumulation in the neutral lipid fraction of the fatty acids prevailing in plant oils. In contrast, the influence of exogenous lipids on the fatty acid composition of bulk fungal phospholipids was insignificant. Nonetheless, a marked increase in the content of membrane lipids and of their phosphatidylethanolamine content was revealed. Presumably, the main mechanism of stimulation of lycopene formation by the oils involves an increase in the solubility of lycopene in the triacylglycerols of the lipid bodies, which is due to an increase in the desaturation degree of their fatty acids. The predominance of linoleic and especially of linolenic fatty acid in plant oils is regarded as a criterion for selecting the oil species for the purpose of intensifying lycopene synthesis.

Key words: Blakeslea trispora, lycopene, lipids

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Biotechnologies aimed at obtaining carotenoids with the use of the mucoraceous fungus *Blakeslea trispora* are based on stimulation of their synthesis via sexual interactions between the heterothallic strains of the fungus. A mixed culture of the (+) and the (–) strain of the fungus forms trisporic acids, lipophilic sex hormones that stimulate carotenoid synthesis [1, 2]. Using this fungus, biotechnologies for obtaining β-carotene and lycopene have been developed [3, 4]. A high antioxidant activity of lycopene provides for its medical applications in respect to diseases associated with oxidative stress including oncological problems. This is exemplified by the use of lycopene for preventing and treating prostate cancer [5–7].

Empirical evidence for the stimulation of carotene formation by supplementing the medium with 3–5% of plant oils has been presented long ago [8]. Theoretically, triacylglycerols, the main component of oils, can be used for the biosynthesis of both carotenoids (involving acetyl-CoA formation resulting from β-oxidation of fatty acids) and the lipids of the fungal mycelium from fatty acids and glycerol (resulting from oil hydrolysis by fungal exolipases).

The life cycle of a fungus in surface culture involves, during the sexual interaction of heterothallic strains, the formation of zygospores containing up to 40% of lipids and 0.5% of carotenoids [9]. Zygospores are not formed in submerged mated culture of heterothallic strains, but vacuolization of mycelium and stimulation of carotenoid and lipid synthesis occur. For example, it was established that the amount of phospholipids synthesized doubled under these conditions [10]. Presumably, this is due to intense formation of lipid bodies coated by a monolayer membrane and predominantly containing tri- and diacylglycerols in which carotenoids are deposited.

Importantly, the stimulatory effect of exogenous trisporic acids and β-ionone, a stimulator of carotenoid synthesis, takes place only in the presence of plant oils in the growth medium [1]. Hence, exogenous oils can (i) be used by the fungus for synthesizing intracellular compounds and obtaining energy and (ii) promote the transfer of the lipophilic compounds (trisporic acids and β-ionone) into the cell. A possible key factor responsible for the dependence of carotenoid synthesis on the presence of exogenous lipids in the medium is the lipophilicity of both trisporic acids and carotenoids per se, with lycopene significantly less soluble in oils than β-carotene.

In view of the wide variety of plant oils, it is mandatory to establish criteria for their selection. The few studies conducted up to now have only demonstrated the influence of oils on the total lipid fraction, which does not contribute to our understanding of the mechanism of their action on carotenoid and lipid synthesis [8, 11]. The data obtained are ambiguous. For instance, it was suggested that stimulation of carotenoid synthesis depends on the total content of (i) oleic ($C_{18:1}$) and linoleic ($C_{18:2}$) acids [8] or (ii) linoleic and linolenic ($C_{18:3}$) acids [11] in oils. Accordingly, the issue concerning the effect of exogenous lip-

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ids on carotenoid synthesis and, in particular, on lycopene formation is of paramount importance.

The goal of this work was to investigate the influence of exogenous lipids on the growth, lycopenegenesis, and lipid composition of the fungus *Blakeslea trispora*.

MATERIALS AND METHODS

The studies were conducted with the T(+) and T(–) strains of *Blakeslea trispora* from the collection of the Winogradsky Institute of Microbiology, Russian Academy of Sciences. The effect of exogenous lipids on lycopene formation was studied in submerged culture. Initially, spore suspensions were used to obtain inocula, i.e., the (+) and (–) cultures in 50 ml of hydrolytic flour medium [12] in 750 ml flasks. Cultivation was carried out for 2 days on a rotary shaker (220 rpm) at 27–28°C. The resulting cultures of the (+) and (–) strains (20% vol/vol) were inoculated at a ratio of 1 : 7 into a 750-ml flask containing 50ml of flour medium [12]. Upon inoculation, 0.005% of 2-amino-6-methylpyridine, a stimulator of lycopene formation, and 5% (wt/vol) of a plant oil were added. The mixed culture was grown for 4 days on a rotary shaker (220 rpm) at 27–28°C.

Olive (Aceites Borges Pont, Spain), linseed (Lyon LLC, Russia, TOR 9141-002-55854031-03), sunflower (Bunge CIS LLC, Russia, State Standard R 52465-2005), cotton (Golden Crown, Uzbekistan), mustard (Aromaty Zhizni, Russia, TOR 9141-020-57023216-04), castor (Tula Pharmaceutical Factory LLC, Russia), and rapeseed (Russian Seeds, Venev Oil Factory Closed Joint-Stock Co., Russia, State Standard 8988-2002) oil were used as exogenous lipids.

According to the Nichols method [13], lipids were extracted from the fungal mycelium with isopropanol twice and thereupon with an isopropanol–chloroform (1 : 1) and an isopropanol–chloroform (1 : 2) mixture. At each stage, the extraction was carried out for 30 min at 70°C. The composition of neutral lipids (NLs) was analyzed by ascending TLC on glass plates with silica gel 60 (Merck, United States). To separate NLs, the following solvent system was used: hexane–diethyl ether–acetic acid (85 : 15 : 1). The NLs were identified using individual markers for mono-, di-, and triacylglycerols, free fatty acids, sterols (ergosterol), and hydrocarbons (Sigma, United States). To analyze phospholipids (PLs) and glycolipids (GLs), the Benning system [14] for two-dimensional TLC was used. The lipids (150–200 μg) were applied to an HPTLC plate with silica gel 60 (Merck, United States). The chromatograms were sprayed with 5% sulfuric acid in ethanol and subsequently heated to 180°C until the stains appeared. To identify PLs, the following individual markers and qualitative tests were used: the ninhydrin test for amino groups, the Dragendorff reagent for choline-containing PLs, and the α-naphthol test for carbohydrates [15]. To identify the sphingolipid moiety of GLs, the saponification test was used [15]. The quantitative analysis of lipids was carried out with the Dens software package (Lenchrom, Russia). The quantities of PLs, sphingolipids, and sterols were assessed using phosphatidylcholine (Sigma), a glycoceramide mixture (Larodan, Sweden), and stigmasterol (Sigma), respectively. The NL ratio was determined with a Sorbphil-M densitometer (Sorbpolymer Closed Joint-Stock Co., Russia).

The fatty acid composition of NLs, PLs, and GLs was determined as methyl esters [15] with a Kristall 5000.1 gas–liquid chromatograph (Chromatek Closed Joint-stock Co., Russia) on an Optima-240 0.25 μm × 60 m × 0.25 mm capillary column (Macherey-Nagel GmbH&Co., Germany) in the 130–240°C temperature range. Identification was carried out using a mixture of individual markers (methyl esters of fatty acids, Supelco 37 Component FAME Mix, United States). Carotenoid composition was assessed using a technique developed by us [16].

The formation of lycopene crystals was used as a criterion of the solubility of lycopene in oils. For this purpose, 0.1% of lycopene was added to the tested oil and completely dissolved at 60°C. Diluted 0.00025 to 0.4% solutions were prepared. They were incubated for 3 days at room temperature until crystals were formed in some of the solutions.

The results were statistically processed using the median (Me) method at $n = 4–6$ [17].

RESULTS AND DISCUSSION

It was established earlier that the lipids of *B. trispora* growing on glucose predominantly contained linoleic ($C_{18:2}$), oleic ($C_{18:1}$), and palmitic ($C_{16:0}$) fatty acid [18]. Therefore, fatty acid composition was used as a criterion for selecting the oils for investigation. Both the oils that were similar to the fungus in fatty acid composition and those differing from it were used. Sunflower and cotton oil predominantly containing linoleic and oleic acid and olive oil whose main fatty acid is oleic acid are similar to the fungus in fatty acid composition. The second group includes linseed oil containing much of polyunsaturated linolenic acid ($C_{18:3}$); mustard oil with linoleic, linolenic, and a number of monoenic acids ($C_{18:1}$, $C_{20:1}$, and $C_{22:1}$); castor oil, the main fatty acid of which is ricinoic acid ($C_{22:1}$-OH); and rapeseed oil dominated by erucic acid ($C_{22:1}$) [19].

By assessing the fatty acid profile of the tested oils, we determined the complete composition of their fatty acids (Table 1). However, it was revealed that rape oil lacked erucic acid, its characteristic component. Therefore, this oil was not used in our subsequent studies. The highest desaturation degree of fatty acids was exhibited by linseed oil (2.12) and the lowest by olive oil (0.96).

Since the fungus was grown in a medium with soybean and corn flour, we assessed the quantities of lipids

Table 1. Fatty acid composition of plant oils

Fatty acids	Fatty acids, % of the total						
	Olive	Linseed	Sunflower	Cotton	Mustard	Castor	Rapeseed
$C_{14:0}$	–	–	–	0.61	–	–	–
$C_{16:0}$	11.04	6.51	6.64	20.76	4.06	1.98	5.04
$C_{16:1}$	0.75	–	–	–	–	–	0.25
$C_{18:0}$	3.01	4.02	2.91	3.15	1.87	1.92	1.76
$C_{18:1}$	71.26	20.22	25.44	20.53	33.27	4.59	59.31
$C_{18:2}$	6.22	14.79	64.63	52.64	27.90	7.40	23.81
$C_{18:3}$	0.504	52.03	–	2.32	10.01	0.57	4.99
$C_{20:0}$	0.37	–	–	–	–	–	0.57
$C_{20:1}$	–	–	–	–	7.35	0.39	1.53
$C_{20:2}$	2.79	2.04	–	–	–	–	2.18
$C_{22:0}$	–	–	0.38	–	–	–	–
$C_{22:1}$	–	–	–	–	15.53	–	0.57
$C_{22:6}$	2.27	0.39	–	–	–	–	–
$C_{22:1}$–OH	–	–	–	–	–	83.16	–
Desaturation degree (CH)	0.96	2.12	1.55	1.33	1.42	1.04	1.29

in these substrates. The assay revealed that only 0.4% (wt/vol) of the lipids were added to the medium with the flour. In all experimental systems, the addition of exogenous oils (5%) to the medium resulted in a two to fivefold and two to threefold increase in biomass yield and lipid amount, respectively, in comparison to the control system without oil (Fig. 1). The influence of oils on lycopene synthesis was more specific because only linseed, cotton, and sunflower oil markedly stimulated lycopene formation (Fig. 2).

Fungal NLs included free fatty acids (FFAs), diacylglycerols (DAGs), triacylglycerols (TAGs), hydrocarbons, sterols, and their esters. It was established that the ratio of acyl-containing NLs changed in the presence of oils in the medium. Linseed, cotton, and sunflower oils that stimulated lycopene formation increased TAG content to 73% of the total, whereas the olive, mustard, and castor oils that stimulated growth but not carotenoid synthesis increased FFA content (Table 2).

A comparative assay of the fatty acid composition of the NL fractions of the fungus and the oils contained in the medium revealed that, in the experimental system with oils of the first group, the fatty acids that prevail in the oils accumulated in the NL fraction. For instance, olive oil contains about 70% of oleic acid (Table 1), and this acid accounted for 52–69% of the total fatty acids in the FFA, DAG, and TAG fractions

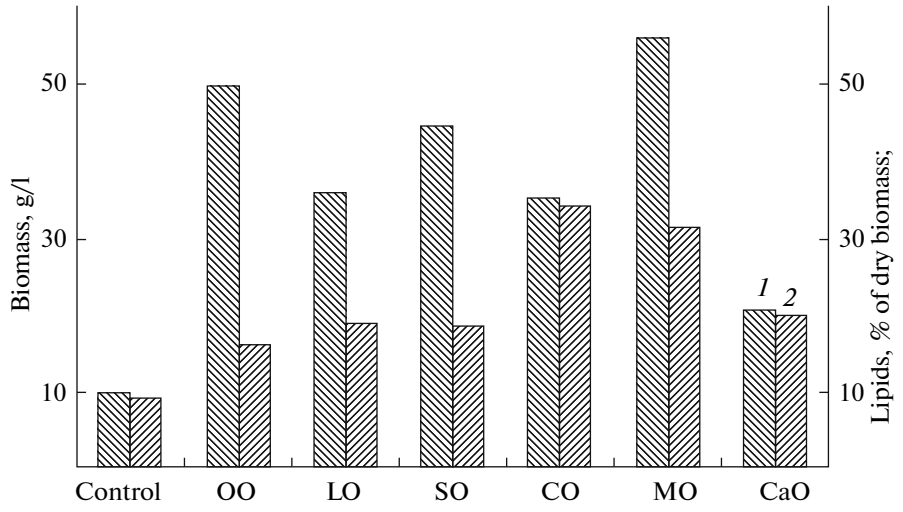

Fig. 1. Influence of exogenous lipids on *B. trispora* growth and lipid accumulation: dry biomass (*1*); lipids (*2*). OO, olive oil; LO, linseed oil; SO, sunflower oil; CO, cotton oil; MO, mustard oil; CaO, castor oil.

(Table 3). The main peculiarity of the effect of cotton and sunflower oil was an increase in the TAG fraction against the background of a decrease in the FFA percentage and significant accumulation of the dominant linoleic acid in the DAG and TAG fractions. Apart from oleic, linoleic, and linolenic acid, mustard oil contains long-chain monoenoic acids $C_{20:1}$ and $C_{22:1}$. However, only trace amounts of these extraneous acids were present in the DAG and TAG fractions, while $C_{22:1}$ fatty acid accumulated in the FFA fraction (60%).The fatty acid composition of the DAGs and the TAGs of the fungus in the presence of mustard oil was similar to that in the control system. The same pattern occurred upon addition of castor oil that contains up to 83% of ricinoic acid ($C_{22:1}$-OH). This acid was not accumulated in any NL fraction, only the FFA fraction contained about 20% of the $C_{22:1}$ fatty acid. Of particular interest is the system with linseed oil, containing up to 52% of linolenic acid (Table 1). Accumulation of linolenic acid (44%), which is not characteristic of the carotenoid-synthesizing (–) strain of the fungus, occurred in the TAG fraction in the presence of linseed oil that exerts the maximum lycopene formation-promoting effect. This points to a paramount importance of linolenic acid for lycopene formation.

The membrane lipids of the fungus consist of phospholipids, sphingolipids (glycoceramides), and sterols (Table 4). All tested oils caused an increase in membrane lipid content (two to nine times higher than in the control) and PL percentage against the background of a decrease in relative sphingolipid content. Unlike the NLs, the main PLs exhibited no significant accumulation of the fatty acids that prevailed in the oils (Table 5). In the systems with linseed and cotton oil that maximally stimulated lycopene formation, a slight increase in linolenic and linoleic acid content

Table 2. Influence of exogenous oils on the composition of the NL fraction

NL fractions	NL, % of the total						
	Without oils	Olive	Linseed	Sunflower	Cotton	Mustard	Castor
TAGs	57.0	65.7	70.1	72.7	73.2	47.5	53.9
FFAs	12.0	31.4	14.5	14.9	9.7	38.3	20.1
DAGs	31.0	2.9	15.4	12.4	17.1	14.2	26.0

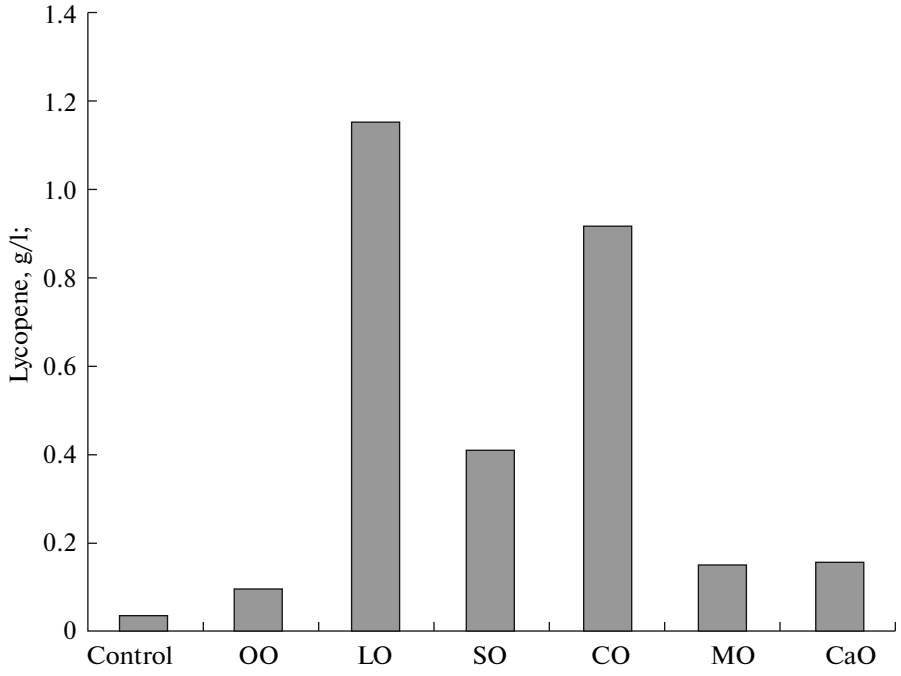

Fig. 2. Influence of exogenous lipids on lycopene formation in *B. trispora*.

was revealed in the phosphatidylethanolamine and phosphatidylcholine fractions, respectively. This resulted in an increase in the desaturation degree of these PLs.

A comparative study concerning the effect of oils on lycopene formation revealed that the strongest effects were produced by oils predominantly containing the $C_{18:2}$ (cotton and sunflower oil) and especially $C_{18:3}$ (linseed oil) fatty acids. These fatty acids accumulated in the TAG and DAG fractions that are the main components of the intracellular lipid bodies where carotenoids are deposited [20]. The analysis of the fatty acid composition demonstrated that neither $C_{18:1} + C_{18:2}$ nor $C_{18:2} + C_{18:3}$ content could act as indices of the stimulation of lycopene formation. The data obtained enabled us to hypothesize that there is a relationship between the composition of the oils added to the medium and their effect on carotenoid synthesis [8, 11]. According to this hypothesis, the key factor is the presence of a large quantity of linoleic and, still more importantly, linolenic acid in the exogenous oils.

The data on the maximum efficiency of oils with high linolenic acid content are of particular interest. The biotechnology of lycopene production we developed involves a mated culture of the (+) and (–) strains at a ratio of 1 : 7, because the (–) strain plays the key role in carotenoid synthesis. It was established earlier [21] that linolenic acid is present in the lipids of the (+) strain. Its percentage can be as high as 20% of the total fatty acid fraction. In contrast, the $C_{18:3}$ acid was not revealed in any of the lipid fractions of the (–) strain of the fungus at any developmental stage [21] even under cold shock [22]. This fact enabled us to suggest a criterion for sex determination in *B. trispora* that is based on the presence/absence of linolenic acids in the lipids. Accumulation of this acid in the TAGs in the presence of linseed oil indicates that this acid promotes lycopene formation in the (–) strain.

It is currently accepted that lipid bodies, apart from deposits of lipophilic reserve substances, are active organelles that contain a number of enzymes involved in lipid metabolism [20]. In this work, we demonstrate that stimulation of lycopene formation is accompanied by an increased content of membrane lipids and triacylglycerols and enhanced desaturation degree of the fatty acids of the NLs. Such changes in the DAG and TAG composition can increase the solubility of the lipophilic lycopene and, accordingly, the capacity of lipid globules. This suggestion is indirectly supported by the fact that the system with linseed oil contained less lipids than that with cotton oil. An increase in the capacity of the lipid granules would enable the fungus to use exogenous lipids, apart from synthesizing intracellular lipids, to form carotenoids and, in particular, lycopene. Assessing the solubility of lycopene in oils revealed that linseed oil exhibits the highest solubility value (0.03%), which is three times lower in sunflower and cotton oil (0.01%). Olive oil has the lowest solubility value (0.005%).

It should be emphasized that the effects of exogenous oils on the acyl chain composition of the main PLs, phosphatidylethanolamines and phosphatidyl-

Table 3. Influence of exogenous lipids on the fatty acid composition of the NLs

Fatty acids	Fatty acids, % of the total																				
	Control, without oil			Olive			Linseed			Sunflower			Cotton			Mustard			Castor		
	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs	DAGs	FFAs	TAGs
$C_{12:0}$	–	–	0.70	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	0.32	–
$C_{14:0}$	0.62	1.71	2.23	1.32	0.54	0.26	–	–	0.23	0.66	1.13	0.29	–	3.41	0.52	0.80	–	–	1.55	0.73	2.37
$C_{14:1}$	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
$C_{15:0}$	–	–	–	–	0.23	–	–	–	0.16	–	–	–	–	1.96	–	–	–	–	–	–	0.42
$C_{16:0}$	18.4	18.4	22.8	16.8	7.56	11.1	11.7	13.4	7.74	9.19	12.5	7.69	16.2	19.8	18.5	12.5	1.59	8.82	14.7	15.0	21.6
$C_{16:1}$	0.32	–	0.27	0.38	–	0.70	–	–	0.21	–	0.16	0.26	–	–	0.50	0.25	–	0.40	1.05	0.21	2.21
$C_{17:0}$	0.26	–	0.98	1.60	0.36	–	1.05	0.83	0.59	0.86	1.42	–	0.97	0.75	–	1.44	–	0.24	0.78	0.33	0.56
$C_{17:1}$	–	–	1.00	–	–	–	–	–	–	0.38	1.46	–	–	–	–	–	–	–	–	–	–
$C_{18:0}$	4.64	16.9	9.40	12.7	12.2	1.58	7.65	19.8	1.32	4.72	21.5	1.4	5.06	20.2	1.77	6.11	6.16	2.35	7.02	10.3	5.18
$C_{18:1}$	25.1	26.5	24.3	52.3	69.2	65.4	35.3	48.6	22.1	25.1	34.4	27.2	28.6	20.4	25.3	41.0	3.74	39.8	22.9	18.1	16.6
$C_{18:2}$	44.9	16.0	32.3	12.1	4.67	16.8	22.0	8.39	23.1	53.1	16.4	59.6	47.5	15.2	50.1	28.9	1.22	36.9	33.5	21.0	39.2
$\tilde{N}_{18:3}$	3.33	–	2.61	0.52	0.62	0.76	22.0	5.68	43.9	1.45	0.62	1.92	1.48	2.40	1.66	4.12	0.77	5.86	4.03	1.41	6.09
$C_{20:0}$	0.39	4.4	0.45	–	1.07	–	–	0.97	–	1.05	1.92	–	–	2.69	–	0.23	2.57	0.19	0.69	1.52	0.87
$C_{20:1}$	0.53	3.32	1.78	–	1.42	0.84	–	2.30	0.20	0.52	2.43	0.35	–	3.59	0.26	1.96	19.0	0.98	2.39	6.04	0.58
$C_{20:2}$	–	–	–	–	–	1.21	–	–	–	–	–	1.25	–	–	–	–	–	0.38	–	0.22	–
$C_{20:3}$	–	–	–	–	–	–	–	–	–	1.64	–	–									
$C_{22:0}$	0.34	4.28	–	–	0.52	–	–	–	–	1.26	–	–	–	2.26	–	0.40	0.86	0.23	–	1.21	1.60
$C_{22:1}$	–	2.84	–	–	0.34	0.44	–	–	–	–	3.14	–	–	5.64	0.34	1.28	60.6	2.58	4.81	20.7	0.77
$C_{22:1}$–OH	–	–	–	–	–	–	–	–	–	–	–	–	–	–	0.62	–	–	–	6.56	0.26	–
$C_{22:2}$	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	0.83	–	–	–	–
$C_{22:6}$	0.49	1.86	1.20	1.31	0.85	0.83	–	–	0.36	–	2.67	–	–	–	–	0.55	–	0.66	–	–	–
$C_{24:0}$	0.58	3.63	–	0.76	0.37	–	–	–	–	–	–	–	–	1.46	0.26	0.43	0.95	0.37	–	1.18	1.92
$C_{24:1}$	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	1.56	0.16	–	1.23	–
Desaturation degree	1.29	0.76	0.98	0.79	0.82	1.06	1.46	0.85	2.01	1.42	0.92	1.55	1.28	0.67	1.32	1.18	0.91	1.40	1.17	0.93	1.17

Table 4. Influence of exogenous oils on the composition of membrane lipids

Exogenous oils	Membrane lipids, % of the total						Total content of membrane lipids, mg/g of dry biomass
	PEAs	PCs	CLs	PAs	GCers	Sts	
Control, without oil	1.79	16.88	5.82	8.34	23.27	43.91	11.82
Olive	13.58	18.49	2.06	1.37	6.49	58.01	22.24
Linseed	17.06	22.71	2.50	5.67	5.71	46.36	63.57
Sunflower	17.65	28.09	Tr.	Tr.	4.92	49.34	65.70
Cotton	22.66	24.97	4.70	8.13	4.65	34.90	83.26
Mustard	18.73	28.40	2.64	3.36	7.68	39.19	96.17
Castor	16.55	29.25	4.09	2.81	3.08	44.22	38.15

Note: Designations: PEAs, phosphatidylethanolamines; PCs, phosphatidylcholines; CLs, cardiolipins; PAs, phosphatidic acids; GCers, glycoceramides; Sts, sterols.

cholines, are relatively weak, unlike the strong influence of the oils on the fatty acid composition of acylglycerols. The dominant fatty acids of the PLs in all experimental variants and in the control were $C_{18:3}$, $C_{18:2}$, and $C_{16:0}$. However, the system with linseed oil was characterized by the presence of a low amount (8.5%) of linolenic acid in the phosphatidylcholine fraction, which resulted in a comparatively high desaturation degree value.

Hence the most probable mechanism of action of exogenous lipids involves accumulation of the dominant polyunsaturated fatty acids in the acylglycerol fraction of the fungus. This results in changes in the desaturation degree of the fatty acids of the acylglycer-

Table 5. Influence of exogenous lipids on the fatty acid composition of the PLs of *B. trispora*

Fatty acids	Fatty acids, % of the total													
	Control, without oil		Olive		Linseed		Sunflower		Cotton		Mustard		Castor	
	PCs	PEAs	PCs	PEAs	PCs	PEAs	PCs	PEAs	PCs	PEAs	PCs	PEAs	PCs	PEAs
$C_{10:0}$	–	4.18	–	–	–		–	–	–	–	–	–	1.47	0.99
$C_{12:0}$	–	4.08	–	–	–	2.07	1.48	1.57	–	–	0.81	–	0.96	–
$C_{14:0}$	1.70	4.04	1.50	1.52	1.07	4.31	6.83	5.97	0.96	–	1.69	1.86	1.80	1.03
$C_{14:1}$	–	–	–	–	–	–	–	–	–	–	0.24	–	–	–
$C_{15:0}$	1.21	1.35	–	–	2.70	–	–	–	0.54	–	0.79	–	1.35	–
$C_{16:0}$	18.62	21.16	15.41	11.22	–	17.64	26.74	22.78	14.53	16.26	16.39	18.72	14.51	6.73
$C_{16:1}$	–	0.44	–	–	–	–	0.59	0.64	–	1.39	0.69	0.30	0.59	–
$C_{17:0}$	2.93	5.94	5.88	10.83	3.37	5.14	1.56	1.88	1.83	1.49	2.78	4.77	13.86	13.68
$C_{17:1}$	–	0.98	0.84	4.05	2.82	–	1.39	2.07	–	3.39	1.64	1.76	3.69	1.49
$C_{18:0}$	13.96	18.07	15.92	29.72	12.65	17.37	13.96	13.99	5.50	13.35	11.78	20.96	12.54	21.25
$C_{18:1}$	30.49	18.60	37.46	25.92	29.64	34.30	29.87	30.70	34.79	33.68	38.28	30.68	15.75	11.66
$C_{18:2}$	23.40	14.13	17.08	9.48	18.66	16.07	16.59	19.29	38.92	22.74	22.17	10.07	23.53	24.61
$C_{18:3}$	0.82	–	–	0.92	8.50	3.10	1.00	1.11	0.86	–	1.55	0.52	1.33	3.17
$C_{20:0}$	0.66	0.76	1.66	1.48	1.35	–	–	–	–	1.45	0.52	0.89	0.62	0.55
$C_{20:1}$	1.18	2.04	2.42	4.88	2.16	–	–	–	0.62	–	0.69	3.49	5.40	5.03
$C_{20:3}$	–	–	1.82	–	–	–	–	–	–	3.41	–	–	–	–
$C_{20:5}$	–	–	–	–	–	–	–	–	–	–	–	–	2.62	1.42
$C_{22:0}$	0.54	0.49	–	–	1.34	–	–	–	–	2.86	–	–	–	–
$C_{22:1}$	1.41	0.90	–	–	11.01	–	–	–	–	–	–	–	–	–
$C_{22:1}$–OH	–	–	–	–	–	–	–	–	–	–	–	–	–	1.12
$C_{22:2}$	–	–	–	–	–	–	–	–	–	–	–	–	–	4.37
$C_{22:6}$	1.00	0.62	–	–	2.13	–	–	–	0.85	–	–	–	–	1.57
$C_{24:0}$	2.09	1.50	–	–	2.62	–	–	–	0.61	–	–	–	–	1.33
$C_{24:1}$	–	0.73	–	–	–	–	–	–	–	–	–	–	–	–
Desaturation degree	0.88	0.56	0.80	0.53	1.21	0.76	0.68	0.75	1.21	0.94	0.90	0.58	0.90	1.03

ols and, accordingly, in an increase in the solubility of lycopene, promoting the accumulation of large amounts of this carotenoid in the lipid bodies.

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