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INFLUENCE of MICROBIAL GROWTH REGULATOR HYDROPTERINE on LACTIC ACID FERMENTATION

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The influence of a novel chemically synthesized growth regulator of microorganisms and plants - hydropterine on the processes of lactic acid fermentation by the cultures of *Lactobacillus delbrueckii* and *Streptococcus bovis* was studied.

Conducted investigations demonstrated high efficiency of mentioned regulator. Being applied in concentrations of 10^{-6} and 10^{-5} g/l of fermentation media hydropterine causes a significant stimulation of both biomass accumulation and lactate production processes. Addition of hydropterine decreases the fermentation period for 11-15% for *L. delbrueckii* and in limits of 18-20% for *S. bovis*.

Ուսումնասիրված է մանրէների և բույսերի նոր, քիմիական եղանակով սինթեզված կարգավորիչի հիդրոպտերինի ազդեցությունը *Lactobacillus delbrueckii* և *Streptococcus bovis* կուլտուրաների կաթնաթթվային խմորման վրա:

Կատարված հետազոտությունները ցույց են տրվել այդ կարգավորիչի բարձր արդյունավետությունը: Հիդրոպտերինը ֆերմենտացիոն միջավայրում 10^{-6} և 10^{-5} գ/լ կոնցենտրացիաներով, զգալիորեն խթանում է արտադրիչների ինչպես բակտերիալ կենսազանգվածի կուտակման, այնպես էլ կաթնաթթվի արտադրության պրոցեսները: Հիդրոպտերինի ավելացումը կրճատում է խմորման պրոցեսի տևողությունը *L. delbrueckii*-ի համար 11-15%-ով, իսկ *S. bovis*-ի 18-20%-ով:

Изучено влияние нового химически синтезированного регулятора роста микроорганизмов и растений - гидроптерина на процессы молочнокислого брожения культур *Lactobacillus delbrueckii* и *Streptococcus bovis*.

Проведённые исследования показали высокую эффективность этого регулятора роста. В концентрациях 10^{-6} и 10^{-5} г/л ферментационной среды гидроптерин приводил к значительной стимуляции процессов как прироста биомассы обоих продуцентов, так и лактатобразования. При добавлении гидроптерина, продолжительность брожения сокращалась на 11-15% для *L. delbrueckii* и на 18-20% для *S. bovis*.

Key words: lactic acid - microbial growth regulator - lactobacteria - hydropterine

Problems related to lactic acid production initiate an increased interest due to the perspectives of its utilization not only as the product for food industry but also a source of novel bio- and photodegradable polymers [4]. The possibilities of use of new efficient lactic acid producers [2, 5], constructions of bioreactors and produc-

tion methods [1, 6, 7, 9], as well as various factors regulating the growth and lactate production of producers have been studied to optimize and intensify lactic acid fermentation processes.

We studied the possibility of stimulation of lactic acid fermentation processes of strains of *Streptococcus bovis* and *Lactobacillus delbrueckii* by a novel growth regulator of microorganisms - hydropterine, synthesized in St. Petersburg State Institute of Technology (Technical University). This substance is easily solved in water and not toxic for human and animals.

Materials and methods. Lactic acid was produced by batch fermentations using as producers the cultures of *S. bovis* or *L. delbrueckii*. For *S. bovis* cultivation corn starch, glucose and inuline were used as carbon sources, while for *L. delbrueckii* - granulated sugar. Yeast extract - 5 g/l, peptone - 5 g/l and CaCO_3 - 50% toward content of starch were added to the fermentation media. Nutritional media were sterilized at excess pressure of 0,5 atmosphere.

Inoculum was produced on the media of following composition (g/l): 20,0, yeast extract - 5,0, peptone - 5,0 and was used after a day of cultivation. Fermentations were carried out in 750 ml fermentation flasks.

Hydropterine was added to the fermentation medium just after inoculation of the bacterial culture.

Concentration of the produced lactate was determined by enzymatic method [8]. The concentration of starch and digestible carbohydrates, as well as remaining sugars was determined by phenol - sulphuric acid method [3].

Results and discussions. The influence of various concentrations of hydropterine on lactic acid production processes of *S. bovis* were studied to determine its optimal concentrations. During lactic acid fermentation on starchy substrates, high concentrations of hydropterine inhibited lactate production, while low concentrations didn't cause any significant unfavorable influence (Table 1). Because the limiting stage of this process is the stage of saccharification of raw cereal starch by α -amylase of the producer culture [3], the assumption was made of the results obtained, that hydropterine does not activate this stage. When the process was carried out on the media containing glucose instead of starch, a valuable stimulation of the process of lactic acid production was observed at the concentrations of hydropterine in 10^{-5} limits of 10^{-6} g/l (Table. 2).

Table 1. The influence of additions of hydropterine on the process of lactic acid fermentation of corn starch by *S. bovis* (starch concentration in the media - 10 %, the ratio of added inoculate to the volume of fermentation media - 10 %.)

Concentration of hydropterine, mg/l	Dynamics of accumulation of lactic acid (g/l). in hours		
	30	56	75
Control, without hydropterine	30	49	61
10^{-4}	24	38	51
10^{-5}	31	51	62
10^{-6}	29	50	60
10^{-7}	29	49	60
10^{-8}	30	50	61

Table 2. The influence of the additions of hydropterine on the lactic acid production process of glucose by *S. bovis* (concentration of glucose in the media - 10 %, ratio of the added inoculum towards (volume of the fermentation media - 10 %)

Concentration of hydropterine, mg/l	Dynamics of accumulation of lactic acid (g/l), in hours		
	28	53	73
Control, without hydropterine	39	55	71
10 ⁻⁴	34	48	61
10 ⁻⁵	54	73	91
10 ⁻⁶	48	58	85
10 ⁻⁷	41	55	74
10 ⁻⁸	38	53	72

The ability of hidropterine in determined optimal concentrations to activate the lactic acid production process on inulin containing media has been also investigated, because the used producer possesses also inulinase activity. The influence of hydropterine was studied for the media with various concentrations of inulin as well as for different ratios of addition of inoculate. In the all cases addition of hydropterine was causing a significant intensification of the process of lactic acid production and decrease of the duration of fermentation (Table 3).

Table 3. The influence of the addition of hydropterine on the process of lactic acid fermentation of inulin by *S. bovis*

Concentration of inulin, g/l	Concentration of added inoculum, %	Concentration of hydropterine, mg/l	Dynamics of accumulation of lactic acid (g/l)		
			28hrs	53hrs	73hrs
75	10	-	43	58	65
		10 ⁻⁶	54	71	71
		10 ⁻⁵	59	71	71
100	10	-	43	57	71
		10 ⁻⁶	53	69	87
		10 ⁻⁵	57	71	93
	15	-	54	71	85
		10 ⁻⁶	59	77	91
		10 ⁻⁵	68	84	92
20	-	75	89		
	10 ⁻⁵	87	91		
150	15	-	83	101	132
		10 ⁻⁵	95	126	139
150	20	-	91	128	138
		10 ⁻⁵	103	139	

The influence of hydropterine on lactic acid production by industrial producer strain of *L. delbrueckii* L-3 was also studied. Obtained results indicate, that

the addition of hydropterine in concentration of 10⁻² g/l to the fermentation media result in intensification of the process for production of lactic acid (Fig. 1). When the influence of this substance on the activity of several key enzymes of *L. delbrueckii* was studied, it was revealed, that the addition of hydropterine to the media at the beginning of the fermentation was increasing the activities of invertase and phosphatase of the producer almost

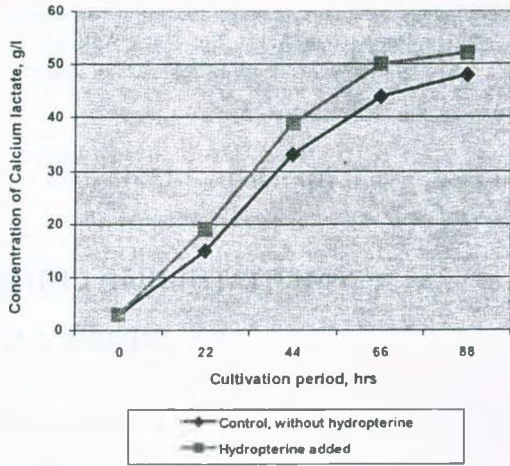


Fig. 1. The dynamics of accumulation of Calcium lactate at fermentation of granulated sugar by *Lactobacillus delbrueckii*.

twice. Fractional addition of hydropterine during the all period

of fermentation appears to be more efficient compared with its only one time addition at the beginning of the process.

The results obtained are demonstrating the efficiency of utilization of hydropterine for intensification of the processes of lactic acid fermentation, while very low concentrations of its stimulating action are proving a prospects and expediency of its utilization.

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