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DISSIMILATORY SULFATE REDUCTION BY VARIOUS *DESULFOVIBRIO* SP. STRAINS OF THE HUMAN INTESTINE

The aim *of this research was to study the dissimilatory sulfate reduction process by various Desulfovibrio sp. strains of the human intestine, such as bacterial growth, sulfate- and lactate usage, production of sulfide and acetate by the strains, and carry out cluster and correlation analyses of this process.* ***Methods.*** *Microbiology methods of the study for bacterial strains cultivation and photometric methods for determination of bacterial biomass and hydrogen sulfide concentration were used, sulfate ions concentration was determined by turbidymetric method, lactate concentration was carried out by lactate dehydrogenase. Acetate ions accumulation by the strains was determined by titration. Using the experimental data, the methods of statistical analysis have been also used.* ***Results.*** *The various Desulfovibrio sp. strains accumulated different biomass for ten days of cultivation in modified Kravtsov-Sorokin's medium. The highest biomass (up to 3.89 g/l) was accumulated by Desulfovibrio sp. strain Vib-7 on the sixth day of cultivation. Clustering of bacterial growth parameters has showed the greatest similarity between strains Desulfovibrio sp. strain Vib-7 and Desulfovibrio sp. strain Vib-9. After using all of the sulfate and lactate from the medium, the bacteria stopped growing and the stationary growth phase began. Clustering of the parameters of sulfate usage has showed that strains Desulfovibrio sp. Vib-1 and Desulfovibrio sp. Vib-2 were combined in one cluster, and the strains Desulfovibrio sp. Vib-7 and Desulfovibrio sp. Vib-9 were in another cluster. The strong correlation between all parameters of dissimilatory sulfate reduction (growth, reduction of sulfate, accumulation of sulfide, use of lactate and accumulation of acetate) by the Desulfovibrio sp. strains has been determined. Thus, the obtained results may be promising for further study, in particular for creating ulcerative colitis models, prediction and prevention of human inflammatory bowel disease.*

Key words: sulfate-reducing bacteria, Desulfovibrio, intestinal microbiocenosis, inflammatory bowel diseases.

Sulfate-reducing bacteria (SRB) of the intestine use different nutrient substances that a human consumes. Human intestinal microbiocenosis is formed by the hundreds of bacterial species and subspecies [2, 4, 5, 9, 17].

It is thought that SRB combined with other infections can cause a variety of diseases including cholecystitis, brain abscesses and abdominal cavity, ulcerative enterocolitis, etc. [5, 10; 13]. The species of *Desulfovibrio* genus are often isolated among all SRB during illness. Bacteria of *Desulfovibrio* genus are also isolated during both mono- and polymicrobial infections of the gastrointestinal tract [6, 7, 14, 15].

It is of vital importance to obtain new strains of SRB from different people while studying the growth of the bacterial strains and the process of dissimilatory sulfate reduction by SRB. The hydrogen sulfide and acetate production by the bacterial strains should also be studied in order to clarify the etiological role of these bacteria in the development of various diseases. The data on the concentration of hydrogen sulfide and acetate produced by the strains, are supposed to help in establishing and assessing a toxicity effect of hydrogen sulfide and acetate on the epithelial cells of the human intestine. Such studies might help in predicting the possibility of appearance of the diseases. It is very important for clinical diagnosis of bowel diseases to get more details on their etiology.

The aim of this research was to study the dissimilatory sulfate reduction process by various *Desulfovibrio* sp. strains of the human intestine, such as bacterial growth, sulfate- and lactate usage, production of sulfide and acetate by the strains, and carry out the cluster and correlation analyses of this process.

Material and methods

The object of the study was sulfate-reducing bacteria of various *Desulfovibrio* sp. strains (SRB Vib-1, SRB Vib-2, SRB Vib-3, SRB Vib-4, SRB Vib-5, SRB Vib-6, SRB Vib-7, SRB Vib-8, SRB Vib-9, SRB Vib-10) obtained from the human large intestine [11].

The bacterial strains were grown in modified Kravtsov-Sorokin's liquid nutrition medium of such composition (g/l): Na_2SO_4 — 0.5; KH_2PO_4 — 0.3; K_2HPO_4 — 0.5; $(NH_4)_2SO_4$ — 0.2; NH_4Cl — 1.0; $CaCl_2 \cdot 6H_2O$ — 0.06; $MgSO_4 \cdot 7H_2O$ — 0.1; $C_3H_5O_3Na$ — 2.0; yeast extract — 1.0; $FeSO_4 \cdot 7H_2O$ — 0.004; sodium citrate$\cdot 2H_2O$ — 0.3. Before bacteria seeding in the medium, 0.05 ml/l of sterile solution of $Na_2S \cdot 9H_2O$ (1%) was added. To provide pH 7.2 of medium, sterile 10N solution of NaOH (0.9 ml/l) was used. The medium was heated in boiling water for 30 min in order to obtain an oxygen-free medium, and cooled to 25 °C. The bacteria were grown for 10 days at 35 °C under anaerobic conditions. The tube was brim-filled with medium and closed by a rubber plug to provide anaerobic conditions.

Accumulation of biomass by various strains of sulfate-reducing bacteria in liquid medium (the medium was without Mohr's salt) was determined by the turbidity of a dilute suspension of cells by the photometric method.

The sulfate ion concentration in the medium was determined by the turbidymetric method after it had been precipitated by barium chloride. To stabilize the suspension, glycerol was used [8].

Hydrogen sulfide in the culture medium was photometrically determined using a spectrophotometer (λ=665 nm, cuvette with optical path 30 mm). The reaction mixture had the following composition: zinc citrate (27.3 mM) — 10 ml; distilled water — 1.98 ml; p-aminodimethylaniline solution (5.5 mM) — 4 ml, and 20 μl of test solution. After 5 min, 1 ml of ferric chloride (0.125 M) was added and methylene blue formation was observed. The concentration of hydrogen sulfide was established by a calibration curve.

Determination of lactate concentration was carried out through a dehydrogenation reaction of lactate by lactate dehydrogenase in the presence of NAD^+, with formation of pyruvate and NADH. For determination of lactate content the following reagents were used: hydrazine–glycine buffer pH 9.0 (glycine — 0.1 M solution containing 0.1 M hydrazine); NAD^+ — 0.03 M solution, pH 6.0; lactate dehydrogenase solution (protein content was about 2 mg/ml). The samples of glycine and hydrazine were dissolved in a small amount of distilled water, pH of 9.0 was maintained by 2 N NaOH solution, then the mixture was diluted by distilled water to 100 ml. Solutions of NAD^+ and lactate dehydrogenase were kept on ice. The content of the tubes was thoroughly mixed and placed inside a thermostat at +25°C for 60 min. After incubation, the samples were cooled and then the optical density of the samples was measured at 340 nm. The quantity of the lactic acid was subsequently calculated [19].

Accumulation of acetate ions by the bacteria cultures during their growth in the medium was determined by titration [1].

The main result of correlation is called Pearson's correlation coefficient (r) [3] was calculated using Excel program.

Using the experimental data, the basic statistical parameters (M — mean, m — standard error, M±m) have been calculated. For the estimation of the reliability between the statistical characteristics, the Student's t-test was used. The difference was reliable when $p>0.95$ [12]. The statistical processing of the results was performed using packet Excel and Origin computer programs. The cluster analysis of parameters of the sulfate reduction was performed using Statistica 6.0 program (Complete Linkage).

Results and Discussion

The results of this research show that all isolated strains were growing actively using sulfate as an electron acceptor and accumulating hydrogen sulfide in the medium. These bacterial strains used lactate as the electron donor; the lactate was actively incompletely oxidized by the bacteria to acetate. The strains of SRB have been accumulating different biomass for ten days of cultivation. The different growth rates of the various strains of

SRB in the modified Kravtsov-Sorokin's medium have been studied. The studied SRB with varying intensity used sulfate and lactate, and produced hydrogen sulfide and acetate. The intense growth of these bacteria depended on the use of sulfate and lactate, and the accumulation of hydrogen sulfide and acetate.

Having used all the sulfate and lactate in the medium, the bacteria stopped growing and the stationary growth phase began. Among all of the isolated bacteria the highest biomass (up to 3.89 g/l) was accumulated by *Desulfovibrio* sp. strain Vib-7 on the sixth day of cultivation. The lowest biomass (up to 3.41 g/l) among vibrios strains was accumulated by *Desulfovibrio* sp. strain Vib-10 on the eighth day of cultivation (fig. 1*A*).

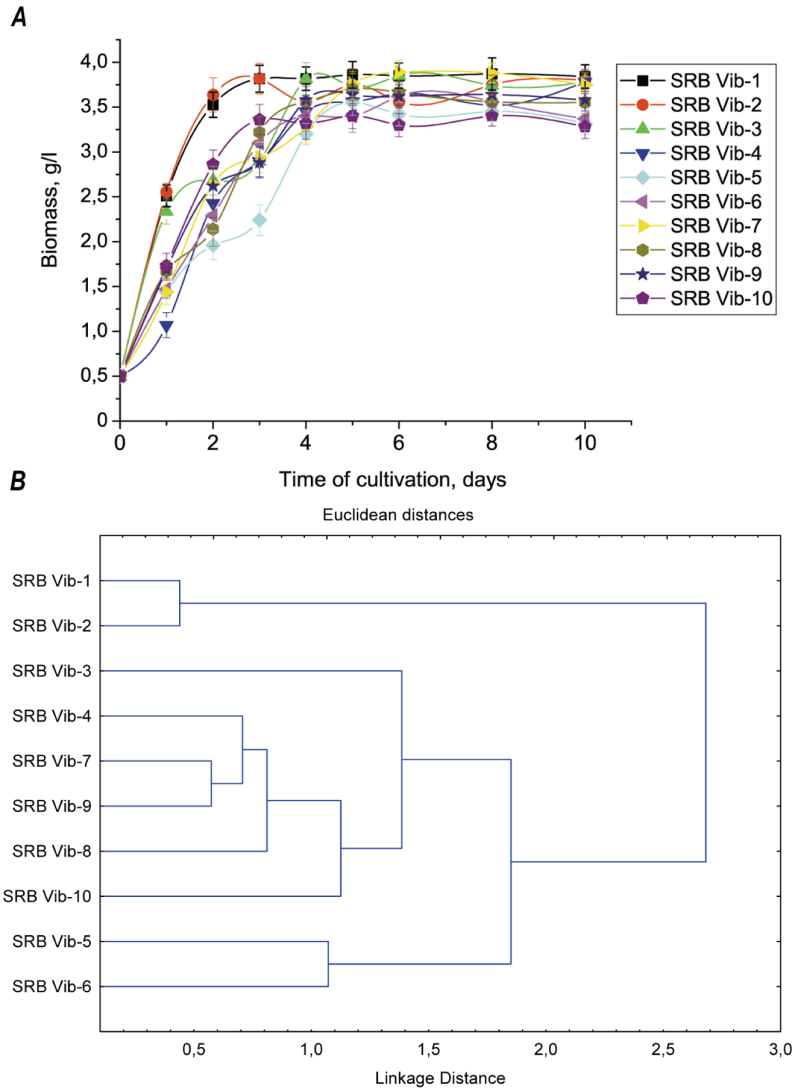

Fig. 1. Growth of the various *Desulfovibrio sp.* strains of the human intestine (A) and dendrogram showing the result of the clustering parameters (B)

As a result of the clustering of the growth parameters there were formed three independent clusters. The dendrogram shows the greatest similarity between strains *Desulfovibrio* sp. strain Vib-7 and *Desulfovibrio* sp. strain Vib-9 by the tested parameter (fig. 1*B*). The strains *Desulfovibrio* sp. Vib-1 and *Desulfovibrio* sp. Vib-2 as well as *Desulfovibrio* sp. Vib-5 and *Desulfovibrio* sp. Vib-6 are the most varied by tested parameter compared with strains *Desulfovibrio* sp. Vib-7 and *Desulfovibrio* sp. Vib-9, and they form two separate isolated clusters.

The studied bacterial strains of *Desulfovibrio* sp. actively reduced the sulfate ions and the bacteria used these ions as an electron acceptor. The intensity and the time of sulfate reduction was different in each of the strains. All of the studied strains fully used sulfate on the sixth day of cultivation (fig. 2*A*).

The parameters of intensity of utilization sulfate by the studied strains are the most similar to each other, allowing combining them in the clusters.

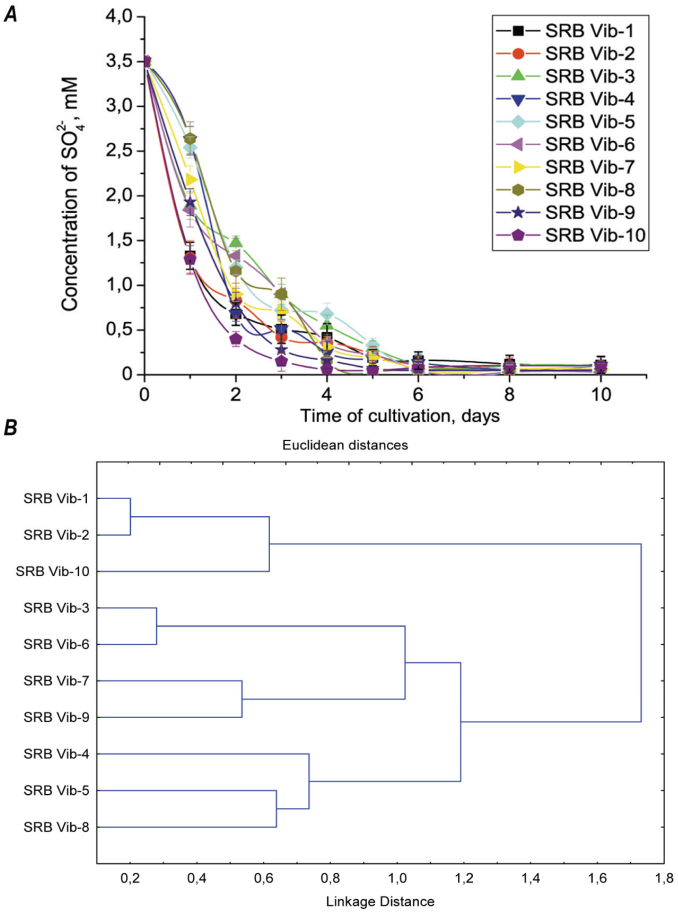

Fig. 2. Sulfate usage by various *Desulfovibrio sp.* strains of the human intestine (A) and the dendrogram showing the result of the clustering parameters (B)

In this case, there is also the formation of three clusters, combining of strains *Desulfovibrio* sp. Vib-1 and *Desulfovibrio* sp. Vib-2 in one cluster, and the strains *Desulfovibrio* sp. Vib-7 and *Desulfovibrio* sp. Vib-9 in another cluster (fig. 2*B*). However, if in this series of the experiments strains *Desulfovibrio* sp. Vib-1 and *Desulfovibrio* sp. Vib-2 still formed stable isolated cluster then the strain *Desulfovibrio* sp. Vib-5 was more similar to the strain *Desulfovibrio* sp. Vib-8, which was not observed in the previous series of experiments.

The highest concentration of hydrogen sulfide (up to 3.23 mM) among the studied strains was produced by bacterial *Desulfovibrio* sp. strain Vib-7 on the eighth day of cultivation; while the bacteria used about 98% of sulfate in the medium, compared to the initial concentration of sulfate. *Desulfovibrio* sp. strain Vib-8 produced hydrogen sulfide in the lowest concentration (up to 2.67 mM) on the sixth day of cultivation compared to all of the studied strains. Under these conditions about 97% of sulfate ions were used in the medium compared to its initial concentration (fig. 3*A*).

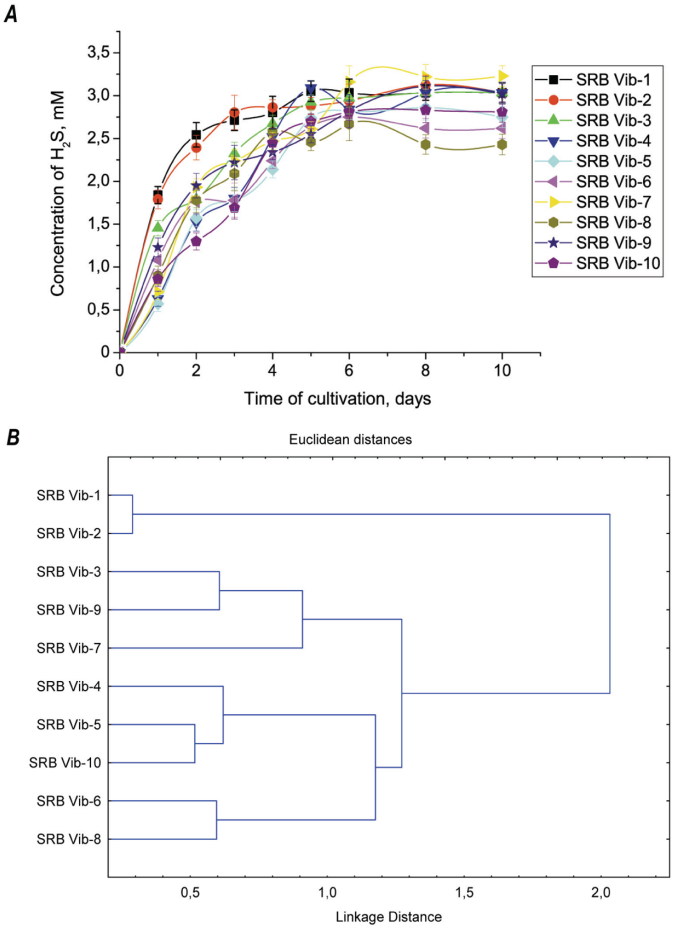

Fig. 3. Production of hydrogen sulfide by the various *Desulfovibrio sp.* strains of the human intestine (A) and the dendrogram showing the result of the clustering parameters (B)

The results clustering of quantitative formation of hydrogen sulfide have shown that *Desulfovibrio* sp. strain Vib-9 with *Desulfovibrio* sp. strain Vib-3, and *Desulfovibrio* sp. strain Vib-7 form one cluster, and the strains *Desulfovibrio* sp. Vib-5 with *Desulfovibrio* sp. Vib-10, and *Desulfovibrio* sp. Vib-4 form the second cluster, as well as *Desulfovibrio* sp. strain Vib-6 and *Desulfovibrio* sp. Vib-8 form the third cluster. *Desulfovibrio* sp. strain Vib-1 and *Desulfovibrio* sp. strain Vib-2 were formed into one stable cluster as in the previous series of the experiments.

An important indicator of the bacterial growth was the presence of organic compounds in the medium. These compounds may simultaneously be a carbon source and an electron donor in the process of dissimilatory sulfate reduction [2, 9, 10].

The results of the studies have showed that the presence of lactate and sulfate in the medium stimulates growth of the studied sulfate-reducing bacteria *Desulfovibrio* sp. various strains. Lactate is an electron donor during dissimilatory sulfate reduction. All studied strains used lactate fully on the sixth day of cultivation, while there were used about 99% of lactate for the dissimilation of sulfate (fig. 4*A*).

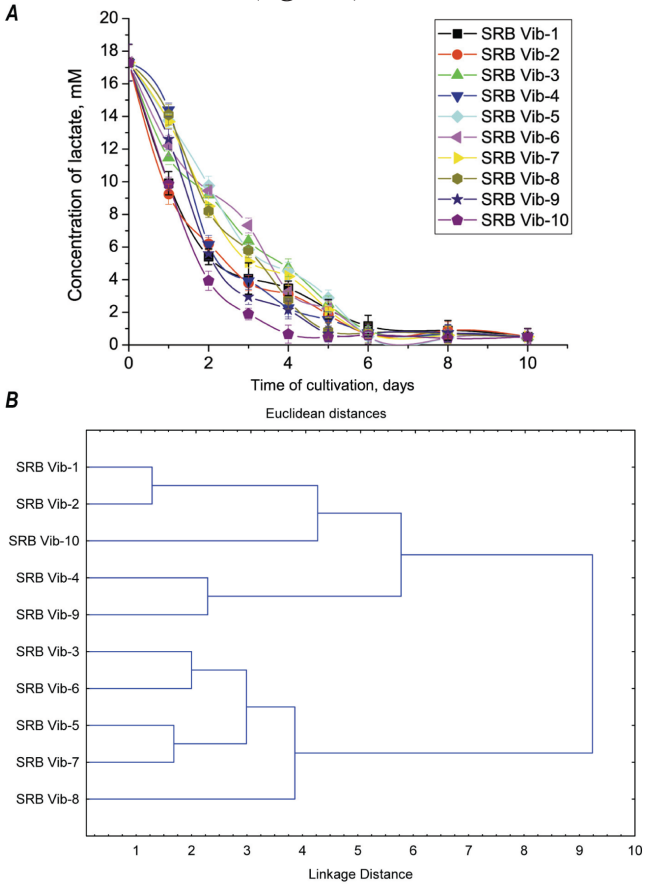

Fig. 4. Lactate usage by the various *Desulfovibrio sp.* strains of the human intestine (A) and the dendrogram showing the result of the clustering parameters (B)

The clustering of the parameters of lactate usage by the studied strains has showed that the strains *Desulfovibrio* sp. Vib-1 with *Desulfovibrio* sp. Vib-9 and *Desulfovibrio* sp. Vib-10, as well as *Desulfovibrio* sp. Vib-4 with *Desulfovibrio* sp. Vib-9 form cluster. Moreover, the *Desulfovibrio* sp. strain Vib-3 with *Desulfovibrio* sp. strain Vib-6 and *Desulfovibrio* sp. strain Vib-7 with *Desulfovibrio* sp. strain Vib-5 as well as *Desulfovibrio* sp. strain Vib-8 form another cluster. In this case, there are formed a clear division into two isolated cluster on the dendrogram (fig. 4*B*).

Among all of the studied strains, *Desulfovibrio* sp. strain Vib-7 produced the highest concentration of acetate ions (up to 15.87 mM) on the fifth day of cultivation; while the bacteria used about 97% of lactate in the medium, compared to its initial concentration of lactate. The *Desulfovibrio* sp. strain Vib-5 produced the lowest concentration (up to 14.36 mM) of acetate ions on the eighth day of cultivation, compared with the vibrio-shaped strains. Under these conditions, the strain used about 98% of lactate in the medium, compared to its initial concentration (fig. 5*A*).

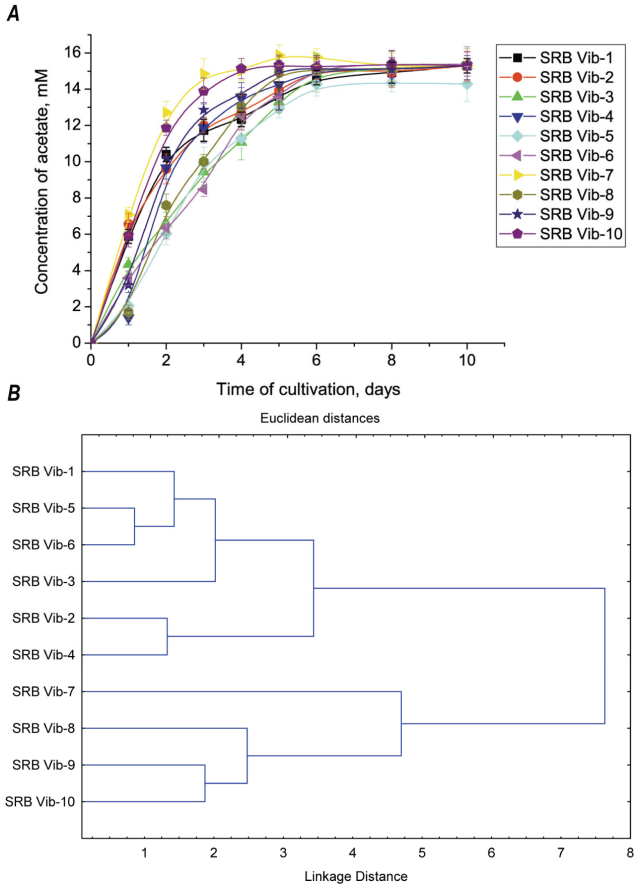

Fig. 5. Acetate production by the various *Desulfovibrio sp.* strains of the human intestine (A) and the dendrogram showing the result of the clustering parameters (B)

The results of the cluster analysis of the parameters of the acetate production by the strains have showed the formation of two separate clusters on the dendrogram (fig. 5*B*). Bacterial strains *Desulfovibrio* sp. Vib-2 with *Desulfovibrio* sp. Vib-4, *Desulfovibrio* sp. strain Vib-6 with *Desulfovibrio* sp. strain Vib-5 and *Desulfovibrio* sp. Vib-3 as well as *Desulfovibrio* sp. Vib-1 form one cluster. The *Desulfovibrio* sp. strain Vib-7 and *Desulfovibrio* sp. strain Vib-8 as well as *Desulfovibrio* sp. strain Vib-9 with *Desulfovibrio* sp. strain Vib-10 form another cluster on the dendrogram by this parameters.

Thus, the results of the studies established that the various studied isolated bacterial *Desulfovibrio* sp. strains performed dissimilatory sulfate reduction intensively. The increased level of sulfates in the intestine can lead to intensive development of *Desulfovibrio* sp. strains and the increase in sulfide and acetate concentrations. Hydrogen sulfide is the main product of metabolism of these strains and can be accumulated in significant quantities in the intestine. Increased sulfide and acetate concentrations can lead to the inhibition of digestive processes in the human intestine. Hydrogen sulfide can also cause cytotoxic and potential carcinogenic effects and the development of intestinal diseases [4, 9, 10, 16, 18]. It is known that hydrogen sulfide affects butyrate oxidation processes and, consequently, can cause damage to the integrity of the epithelial barrier cells and inflammation of the colon epithelium [13, 14, 15]. It is known from the literature that *Desulfovibrio* sp. bacteria cause bloody diarrhea, weight loss and anorexia in animals and human. Under these conditions, epithelial hyperplasia, abscesses and inflammatory infiltrates can occur [10, 14, 18].

The correlation coefficients (r) between the parameters of dissimilatory sulfate reduction by the *Desulfovibrio* sp. strains were defined (Table). Between biomass and sulfate ($r = -0.975$); biomass and lactate ($r = -0.974$); sulfate and sulfide ($r = -0.97$); sulfate and acetate ($r = -0.844$); lactate and acetate ($r = -0.97$); and lactate and sulfide ($r = -0.976$) a strong inversely negative correlation has been estimated.

Between biomass and sulfide ($r = +0.969$); biomass and acetate ($r = +0.85$); lactate and sulfate ($r = +0.982$); acetate and sulfide ($r = +0.894$) a strong positive correlation has been estimated.

The correlation coefficient ranges from -1.0 to +1.0. The closer r is to +1 or -1, more closely the two variables are related. If r is close to 0, it means there is no relationship between the variables. If r is positive, it means that as one variable gets larger the other gets larger. If r is negative it means that as one gets larger, the other gets smaller (often called an «inverse» correlation). While the correlation coefficients are normally reported as $r =$ (a value between -1 and +1), squaring them makes then easier to understand. The values between 0.7 and 1.0 (-0.7 and -1.0) indicate a strong positive (negative) linear relationship via a firm linear rule [3].

Table

Correlation coefficients (r) between dissimilatory sulfate reduction parameters by the various *Desulfovibrio sp.* strains

SRB Vib-1					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.991	0.993	-0.969	0.671
Sulfate	-0.991	1	-0.999	0.979	-0.729
Sulfide	0.993	-0.999	1	-0.983	0.731
Lactate	-0.969	0.979	-0.983	1	-0.832
Acetate	0.671	-0.729	0.731	-0.832	1

SRB Vib-2					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.977	0.98	-0.941	0.698
Sulfate	-0.977	1	-0.997	0.983	-0.803
Sulfide	0.98	-0.997	1	-0.984	0.808
Lactate	-0.941	0.983	-0.984	1	-0.89
Acetate	0.698	-0.803	0.808	-0.89	1

SRB Vib-3					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.986	0.988	-0.957	0.864
Sulfate	-0.986	1	-0.999	0.987	-0.897
Sulfide	0.988	-0.999	1	-0.985	0.896
Lactate	-0.957	0.987	-0.985	1	-0.95
Acetate	0.864	-0.897	0.896	-0.95	1

SRB Vib-4					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.978	0.98	-0.996	0.9
Sulfate	-0.978	1	-0.938	0.99	-0.825
Sulfide	0.98	-0.938	1	-0.97	0.939
Lactate	-0.996	0.99	-0.97	1	-0.889
Acetate	0.9	-0.825	0.939	-0.889	1

SRB Vib-5					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.954	0.98	-0.969	0.891
Sulfate	-0.954	1	-0.982	0.979	-0.848
Sulfide	0.98	-0.982	1	-0.988	0.916
Lactate	-0.969	0.979	-0.988	1	-0.921
Acetate	0.891	-0.848	0.916	-0.921	1

SRB Vib-6					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.943	0.932	-0.957	0.871
Sulfate	-0.943	1	-0.99	0.978	-0.88
Sulfide	0.932	-0.99	1	-0.981	0.899
Lactate	-0.957	0.978	-0.981	1	-0.947
Acetate	0.871	-0.88	0.899	-0.947	1

SRB Vib-7					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.988	0.988	-0.993	0.867
Sulfate	-0.988	1	-0.975	0.972	-0.81
Sulfide	0.988	-0.975	1	-0.993	0.9
Lactate	-0.993	0.972	-0.993	1	-0.897
Acetate	0.867	-0.81	0.9	-0.897	1

SRB Vib-8					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.976	0.981	-0.975	0.934
Sulfate	-0.976	1	-0.99	0.991	-0.934
Sulfide	0.981	-0.99	1	-0.972	0.912
Lactate	-0.975	0.991	-0.972	1	-0.966
Acetate	0.934	-0.934	0.912	-0.966	1

SRB Vib-9					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.983	0.972	-0.988	0.924
Sulfate	-0.983	1	-0.968	0.987	-0.883
Sulfide	0.972	-0.968	1	-0.972	0.948
Lactate	-0.988	0.987	-0.972	1	-0.924
Acetate	0.924	-0.883	0.948	-0.924	1

SRB Vib-10					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.978	0.9	-0.996	0.886
Sulfate	-0.978	1	-0.862	0.979	-0.827
Sulfide	0.9	-0.862	1	-0.93	0.99
Lactate	-0.996	0.979	-0.93	1	-0.91
Acetate	0.886	-0.827	0.99	-0.91	1

Total correlation coefficients for all SRB Vib-1-10 bacterial strains					
	Biomass	Sulfate	Sulfide	Lactate	Acetate
Biomass	1	-0.975	0.969	-0.974	0.85
Sulfate	-0.975	1	-0.97	0.982	-0.844
Sulfide	0.969	-0.97	1	-0.976	0.894
Lactate	-0.974	0.982	-0.976	1	-0.912
Acetate	0.85	-0.844	0.894	-0.912	1

Taking into consideration all of the obtained results: the studies of bacterial *Desulfovibrio* sp. strains growth in the medium, their sulfate and lactate usage, the production of hydrogen sulfide and acetate by the strains, and cluster and correlation analyses of the parameters of the growth by the various *Desulfovibrio* sp. strains, the isolated bacteria may cause various human intestinal diseases and inflammatory bowel processes. Therefore these bacteria are quite interesting and promising for further studies.

Thus, the intense growth of various *Desulfovibrio* sp. strains perhaps depends on the use of sulfate and lactate and the accumulation of hydrogen sulfide and acetate. Having used all of the sulfate and lactate in the medium, the bacteria stopped growing and the stationary growth phase began. Among all of the isolated bacterial strains the highest biomass (up to 3.89 g/l) was accumulated by the *Desulfovibrio* sp. strain Vib-7. Clustering of bacterial growth parameters has showed the greatest similarity between strains *Desulfovibrio* sp. strain Vib-7 and *Desulfovibrio* sp. strain Vib-9 by the tested parameter.

The studied bacterial strains of *Desulfovibrio* genus actively reduce sulfate ions and use these ions as an electron acceptor. The strains used sulfate fully on the sixth day of cultivation. Clustering of the parameters of sulfate usage has showed that the strains *Desulfovibrio* sp. Vib-1 and *Desulfovibrio* sp. Vib-2 were combined in one cluster, and the strains *Desulfovibrio* sp. Vib-7 and *Desulfovibrio* sp. Vib-9 were in another cluster. The highest concentrations of hydrogen sulfide (up to 3.23 mM) and acetate ions (up to 15.87 mM) among all the studied strains were produced by the bacteria *Desulfovibrio* sp. strain Vib-7. The strong correlation between parameters of dissimilatory sulfate reduction by the *Desulfovibrio* sp. various strains has been estimated.

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ДИСИМІЛЯЦІЙНЕ ВІДНОВЛЕННЯ СУЛЬФАТІВ РІЗНИМИ ШТАМАМИ *DESULFOVIBRIO* SP. КИШЕЧНИКА ЛЮДИНИ

Реферат

Метою даного дослідження було вивчити процес дисиміляційного відновлення сульфату різними штамами *Desulfovibrio* sp. кишечника людини, зокрема ріст бактерій, використання ними сульфату і лактату, утворення гідроген сульфіду і ацетату, а також виконати кластерний та кореляційний аналіз цього процесу. **Методи.** Мікробіологічні методи досліджень використані під час культивування штамів бактерій,

фотометричні для визначення бактеріальної біомаси та концентрації гідроген сульфіду; концентрацію іонів сульфату визначено турбідометричним методом, а кількість лактату встановлено за лактатдегідрогеназою. Продукування штамами ацетат іонів визначено методом титрування. На основі експериментальних даних також проведено статистичний аналіз отриманих результатів. **Результати.** Різні штами *Desulfovibrio* sp. накопичували різну біомасу упродовж 10 днів культивування у модифікованому середовищі Кравцова-Сорокіна. Найбільшу біомасу (до 3,89 г/л) накопичували бактерії *Desulfovibrio* sp. штам Vib-7 на шостий день культивування. Кластеризація параметрів бактеріального росту показала найбільшу подібність між штамами *Desulfovibrio* sp. штам Vib-7 і *Desulfovibrio* sp. штам Vib-9. Використавши сульфат і лактат середовища, бактерії переставали рости і виходили на стаціонарну фазу росту. Кластеризація параметрів використання сульфату показала, що штами *Desulfovibrio* sp. Vib-1 і *Desulfovibrio* sp. Vib-2 були об'єднані в один кластер, а штами *Desulfovibrio* sp. Vib-7 і *Desulfovibrio* sp. Vib-9 були віднесені до іншого кластеру. Встановлено високий ступінь кореляції між параметрами дисиміляційного відновлення сульфатів (ростом, відновленням сульфату, нагромадженням сірководню, використанням лактату і утворенням ацетату) штамами *Desulfovibrio* sp. Отже, отримані результати можуть бути перспективними для подальших досліджень, зокрема для створення моделей виразкових колітів, прогнозування і попередження запальних захворювань кишечника людини.

Ключові слова: сульфатвідновлювальні бактерії, *Desulfovibrio*, мікробіоценоз кишечника, запальні захворювання кишечника.

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ДИССИМИЛЯТИВНОЕ ВОССТАНОВЛЕНИЕ СУЛЬФАТОВ РАЗЛИЧНЫМИ ШТАММАМИ *DESULFOVIBRIO* SP. КИШЕЧНИКА ЧЕЛОВЕКА

Реферат

Целью данного исследования было изучить процесс диссимиляционного восстановления сульфата разными штаммами *Desulfovibrio* sp. кишечника человека, в частности рост бактерий, использование ими сульфата и лактата, накопление гидроген сульфида и ацетата, а также провести кластерный и корреляционный анализ этого процесса. **Методы.** Микробиологические методы исследований использованы при культивировании штаммов бактерий, фотометрические для

определения бактериальной биомассы и концентрации гидроген сульфида; концентрацию ионов сульфата определено турбидометрическим методом, а количество лактата установлено по лактатдегидрогеназе. Продуцирование штаммами ацетат ионов определено методом титрования. На основе экспериментальных данных также проведен статистический анализ полученных результатов. **Результаты.** Различные штаммы *Desulfovibrio* sp. накапливали различную биомассу в течение 10 дней культивирования в модифицированной среде Кравцова-Сорокина. Наибольшую биомассу (до 3,89 г/л) накапливали бактерии *Desulfovibrio* sp. штамм Vib-7 на шестой день культивирования. Кластеризация параметров бактериального роста показала наибольшее сходство между штаммами *Desulfovibrio* sp. штамм Vib-7 и *Desulfovibrio* sp. штамм Vib-9. Использовав сульфат и лактат со среды, бактерии прекращали расти и выходили на стационарную фазу роста. Кластеризация параметров использования сульфата показала, что штаммы *Desulfovibrio* sp. Vib-1 и *Desulfovibrio* sp. Vib-2 были объединены в один кластер, а штаммы *Desulfovibrio* sp. Vib-7 и *Desulfovibrio* sp. Vib-9 были отнесены к другому кластеру. Установлена высокая корреляционная зависимость между параметрами диссимиляционного восстановления сульфатов (ростом, восстановлением сульфата, накоплением сероводорода, использованием лактата и образованием ацетата) штаммами *Desulfovibrio* sp. Таким образом, полученные результаты могут быть перспективными для дальнейших исследований, в частности для создания моделей язвенных колитов, прогнозирования и предупреждения воспалительных заболеваний кишечника человека.

Ключевые слова: сульфат-восстановительные бактерии, *Desulfovibrio*, микробиоценоз кишечника, воспалительные заболевания кишечника.

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