

Influence of potassium dichromate on the reduction of sulfur, nitrate and nitrite ions by bacteria *Desulfuromonas* sp.

O. M. Moroz, S. O. Hnatush, H. V. Yavorska, G. I. Zvir, O. V. Tarabas

Ivan Franko National University of Lviv, Lviv, Ukraine

Article info

Received 11.05.2022
Received in revised form
02.06.2022
Accepted 04.06.2022

Ivan Franko National
University of Lviv,
Hrushevskyi st., 4,
Lviv, 79005, Ukraine.
Tel.: +38-067-811-86-44.
E-mail:
oksana.moroz@lnu.edu.ua

Moroz, O. M., Hnatush, S. O., Yavorska, H. V., Zvir, G. I., & Tarabas, O. V. (2022). Influence of potassium dichromate on the reduction of sulfur, nitrate and nitrite ions by bacteria *Desulfuromonas* sp. *Regulatory Mechanisms in Biosystems*, 13(2), 153–167. doi:10.15421/022220

This article presents the regularities of reduction of sulfur, nitrate and nitrite ions by sulfur reducing bacteria *Desulfuromonas* sp., which were isolated from the water of the man-made Yavorivske Lake (Lviv Region, Ukraine), under the influence of potassium dichromate. This bacteria in the process of anaerobic respiration can use and reduce different electron acceptors, such as sulfur, nitrates, nitrites, oxidized forms of heavy metals, in particular, hexavalent chromium. Technogenically altered ecotopes are characterized by complex pollution, so several electron acceptors are available to bacteria at the same time. Strains of microorganisms isolated from such ecotopes are adapted to unfavourable conditions and therefore have high biotechnological potential. The purpose of this work was to investigate the regularities of elemental sulfur, nitrate or nitrite ion usage by sulfidogenic bacteria of *Desulfuromonas* genus in conditions of simultaneous presence in the medium of another electron acceptor – Cr(VI), to establish the succession of reduction of electron acceptors by strains of these bacteria and to evaluate the efficiency of their possible application in technologies of complex purification of the environment from metal compounds and other inorganic toxicants. Bacteria were grown under anaerobic conditions in Kravtsov-Sorokin medium without SO_4^{2-} and without Mohr's salt for 10 days. To study the efficiency of sulfur, nitrate or nitrite ions' reduction at simultaneous presence in the medium of Cr(VI) bacteria were sown in media with elemental sulfur, NaNO_3 , NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final S^0 , NO_3^- , NO_2^- or Cr(VI) concentration in the medium of 3.47 (concentration of SO_4^{2-} in medium of standard composition) or 1.74, 3.47, 5.21, 6.94 and 10.41 mM. Biomass was determined by the turbidimetric method, and the concentrations of nitrate, nitrite, ammonium ions, hydrogen sulfide, Cr(VI), Cr(III) in cultural liquid were determined spectrophotometrically. It has been established that Cr(VI) inhibits the biomass accumulation and hydrogen sulfide production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the medium of 3.47 mM S^0 and 1.74–10.41 mM Cr(VI). In the medium with the same initial content (3.47 mM) of S^0 and Cr(VI) bacteria produced Cr(III) at concentrations 3.3–3.4 times higher than that of hydrogen sulfide. It has been shown that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits biomass accumulation, nitrate ions' reduction and ammonium ions' production by bacteria after simultaneous addition into the medium of 3.47 mM NO_3^- and 1.74–10.41 mM Cr(VI) or 1.74–10.41 mM NO_3^- and 3.47 mM Cr(VI). In the medium with the same initial content (3.47 mM) of NO_3^- and Cr(VI) bacteria reduced up to 1.2 times more nitrate ions than Cr(VI) with the production of ammonium ions at concentrations the same times higher than those of Cr(III). It has been established that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits biomass accumulation, nitrite ions' reduction and ammonium ions' production by bacteria after simultaneous addition into the medium of 3.47 mM NO_2^- and 1.74–10.41 mM Cr(VI) or 1.74–10.41 mM NO_2^- and 3.47 mM Cr(VI). In the medium with the same initial content of (3.47 mM) NO_2^- and Cr(VI) the reduction of Cr(VI) by bacteria was only slightly, up to 1.1 times, lower than the reduction of nitrite ions, almost the same concentrations of trivalent chromium and ammonium ions were detected in the cultural liquid. The processes of nitrate and nitrite reduction carried out by bacteria of *Desulfuromonas* genus were revealed to be less sensitive to the negative influence of sodium dichromate, as compared with the process of sulfur reduction, because in the media with the same initial content (3.47 mM) of NO_3^- or NO_2^- and Cr(VI) bacteria produced 1.1–1.2 times more NH_4^+ than Cr(III), but in the medium with the same initial content (3.47 mM) of S^0 and Cr(VI) bacteria produced over than three times more Cr(III) than hydrogen sulfide. Our data allow us to conclude that bacteria of *Desulfuromonas* genus, the investigated strains of which are adapted to high concentrations (up to 10.41 mM) of inorganic toxicants, play an important role in the geochemical cycles of sulfur, nitrogen and chromium in aquatic environments that have been under anthropogenic influence.

Keywords: *Desulfuromonas* sp.; sulfur; nitrate ions; nitrite ions; hexavalent chromium; electron acceptors.

Introduction

Chromium is one of the most common environmental contaminants. Chromium is routinely discharged to the environment from effluents of multiple industrial processes (chrome plating, dye manufacturing, the textile industry, the aircraft industry, leather tanning, wood preservation, mud drilling, steel, automobile manufacturing, military defense applications). Chromates, dichromates, chromic acid, chromic sulfate and chromic oxides are examples of industrially relevant chromium compounds. These chromium compounds are generally produced from the mining and

treatment of chromite ore. In addition to mining and industrial activities, natural rocks such as ultramafic and mafic rocks, volcanic eruptions, forest fires, and weathering are also a geogenic source of Cr(VI) in groundwater (Viti et al., 2014; Kazakis et al., 2015). Cr has high redox potential, and it can exist in several oxidation states, the most stable of which are Cr(III) and Cr(VI). They differ in chemical characteristics and biological effects (Sharma et al., 2022). Cr(VI) pollution has become one of the world's most serious environmental concerns due to its non-biodegradability and long persistence in the soil and water and highly toxicity to humans and wildlife. Cr(VI) is classified as a group 1 carcinogen by the World Health

Organization, the maximum allowed concentration of chromium in drinking water is set at 50 µg/L by the drinking water guideline (Sharma et al., 2022). Physico-chemical techniques currently used for Cr(VI) removal (adsorption, chemical precipitation, reverse osmosis, ion exchange, electrocoagulation, membrane separation, electrodialysis) are not environmentally friendly and use a large number of chemicals (Fathima et al., 2005). Bioremediation (bioaccumulation, biotransformation, biosorption, bioleaching) is often the preferred method to deal with Cr contaminated sites, because it is an eco-friendly and cost-effective technology. A variety of fungal and bacterial species (*Aspergillus* sp., *Streptomyces rimosus*, *Actinomyces* sp., *Streptomyces griseus*, *Chelatococcus daeguensis*, *Pseudomonas alcaliphila* NEWG-2, *Bacillus* sp., *Pleurotus ostreatus*, *Scenedesmus* sp.) showed promise in removing Cr(VI) from industrial effluent (Poopal & Laxman, 2009; Sharma et al., 2022). Representatives of the sulfidogenic microbiota, isolated from contaminated environments and adapted to stress factors, also are suitable for use in the technologies of purification of different substrates from chromium compounds and other toxicants (Moroz et al., 2014; Kuznetsov et al., 2015; Moroz et al., 2018; Teng et al., 2019).

Cr(III) is relatively insoluble under natural conditions and therefore less toxic than Cr(VI) (Sobol & Schiestl, 2012; Viti et al., 2014). Despite the fact that Cr(III) is an essential oligoelement for humans (Joutey et al., 2015), at high concentrations it affects DNA replication, causes mutagenesis, and alters the structure and activity of enzymes, reacting with their carboxyl and thiol groups in cells (Viti et al., 2014). The hexavalent form of Cr is soluble, carcinogenic, genotoxic, and mutagenic for living organisms (Viti et al., 2014; Liang et al., 2021). In bacteria cells Cr(VI) modifies the expression of genes whose products are involved in transport and metabolism of carbohydrates, amino acid conversion, production and use of energy in the form of ATP (Viti et al., 2014). Cr(VI) penetrate the cell membrane of *Shewanella oneidensis* MR-1, *Pseudomonas putida* F1, *Cupriavidus metallidurans* CH34, *Arthrobacter* sp. FB24 by the ABC sulfate transporter system (Aguilar-Barajas et al., 2011; Hoffmann et al., 2017). In cytoplasm it interacts with intracellular reductants (amino acids, nucleotides, sugars, organic acids, glutathione, flavoenzymes, vitamins) and generates chemically active intermediates Cr(V) or Cr(IV), Cr(III) as the end product, free radicals and can cause oxidative stress (Sobol & Schiestl, 2012; Viti et al., 2014; Hnatysh & Maslovska, 2018). Reduction of Cr(VI) to Cr(V) is combined with the formation of H₂O₂. The interaction of Cr(V) with H₂O₂ leads to the formation of hydroxyl radicals. Probably, the genotoxicity and toxicity of Cr(VI) is due to damage of DNA and proteins, respectively, caused by oxygen radicals, formed as a result of its reduction (Sobol & Schiestl, 2012).

There are several Cr-resistance mechanisms that are displayed by microorganisms. These include active efflux of Cr compounds, metabolic reduction of Cr(VI) to Cr(III), activation of enzymes involved in the detoxification of active oxygen forms and repair of DNA damage, and either intercellular or extracellular precipitation of Cr(VI) (Belchik et al., 2011; Richter et al., 2012; Joutey et al., 2015). Determinants of chromate and dichromate resistance (genes encoding proteins involved in their transport across the membrane) in isolates of Enterobacteriaceae are localized in chromosomal or plasmid DNA (Caballero-Flores et al., 2011). Microbial Cr(VI) removal typically involves three stages: binding of chromium to the cell surface, translocation of chromium into the cell, and reduction of Cr(VI) to Cr(III). Cr(VI) reduction by microorganisms may proceed on the cell surface, outside the cell, or intracellularly, either directly via chromate reductase enzymes, or indirectly via metabolite reduction of Cr(VI) (Sharma et al., 2022). Membrane-bound metal reductases in gram-negative bacteria are associated with the outer side of the membrane to reduction metal ions outside the cell (Lengeler et al., 2005; Richter et al., 2012). For bioaccumulation live biomass uses cellular energy to transport hexavalent chromium through the cell membrane. The uptake of chromium ions is a biphasic process. The primary step is known as biosorption, a metabolic energy independent process. Potentially hazardous heavy metal ions link themselves (by mechanisms of ion exchange, complexation, chelation, adsorption, microprecipitation) to the cell's surface binding sites (phosphates, carboxyl, imidazole, amino, hydroxyl moieties, thioether, sulfate, phenol, amine, sulfhydryl groups). Thereafter, bioaccumulation occurs, but is much slower, and is dependent on cell metabolic activi-

ty. The metal-ligand combination that develops on the cell surface is subsequently transported inside by transporter protein. In addition, intracellularly transported complexes interact with metal-binding proteins such as phytochelatin and metallothionein, causing precipitation, methylation, and other reactions (Joutey et al., 2015; Sharma et al., 2022). Most representatives of sulfidogenic bacteria have a non-specific metal reductase enzyme system that allows them to use compounds of Cr(VI), Fe(III), Mn(IV), U(VI), Cu(II) and other metals as electron acceptors of anaerobic respiration (Kozlova et al., 2008). Soluble and insoluble metal compounds are reduced outside the cells by a system of membrane-bound metal reductases (multi-heme c-type cytochromes) (Gescher & Kappler, 2012; Richter et al., 2012; Breuer et al., 2015), therefore electrons are released into the media, allowing these exoelectrogenic anaerobic bacteria to be used in the microbial fuel cells as the anode biocatalysts (Fitzgerald et al., 2013; Vasylyv et al., 2016; Prokhorova et al., 2017; Simonte et al., 2017). Cytochromes MtrA, MtrB, MtrC and OmcA of *Shewanella oneidensis* MR-1 are involved in dissimilatory reduction of oxidized forms of metals, and it was found that MtrC and OmcA are terminal reductases of extracellular Cr(VI) reduction (Belchik et al., 2011; Jing et al., 2020). Three-, tetra- and deca-heme c-type cytochromes in *Shewanella frigidimarina*, *Desulfovibrio vulgaris*, *Desulfuromonas acetoxidans* are localized between the inner and outer membranes and in the periplasm, through which electrons from the cytoplasm from the reactions of organic compounds oxidation are transferred to the outside of the cells, where the metal ions are reduced (Gescher & Kappler, 2012). Bacteria of the *Desulfuromonas* genus oxidize organic substrates using metals with variable valence as electron acceptors (Moroz et al., 2014; Moroz et al., 2016), reduce and transform them into non-toxic or less toxic for living organisms forms (Vasylyv et al., 2011; Bilyy et al., 2014; Maslovska et al., 2015).

Obligate anaerobic eubacteria *Desulfuromonas acetoxidans*, *Desulfurella acetivorans*, *Wolinella succinogenes*, *Desulfovibrio gigas*, *Desulfomicrobium* sp. can carry out sulfur respiration with the formation of hydrogen sulfide (Kozlova et al., 2008). Bacteria of the *Desulfuromonas* genus use elemental or polysulfide sulfur, nitrates, nitrites, L-malate, fumarate, tri- or tetrachlorethylene, oxidized forms of heavy metals (Cr(VI), Fe(III), Mn(IV), Cu(II)) as electron acceptors (Sung et al., 2003; Kuever et al., 2005; Moroz et al., 2014; An & Picarda, 2015; Hnatysh et al., 2018), oxidizing H₂ or the simple organic compounds to CO₂ (Roden & Lovley, 1993; Hedderich et al., 1999; Vasylyv et al., 2015). In *Wolinella succinogenes* polysulfide reduction occurs in the periplasm, using hydrogen and electron transport chain, which includes hydrogenase and polysulfide reductase. These enzymes are localized in the cytoplasmic membrane and their substrate-binding sites are oriented to the periplasm. Polysulfide reductase contains molybdopterine guanine dinucleotide and Fe-S protein and bound with hydrogenase by cytochromes or quinones (Hedderich et al., 1999; Lengeler et al., 2005). Sulfidogenic bacteria attract attention of biotechnologists as potential agents of purification of contaminated by metal compounds environments, because as a result of interaction H₂S with divalent metal ions their insoluble sulfides are formed and thus they are removed from the natural cycle of elements (Kiran et al., 2017; Moroz et al., 2018). These bacteria reduce nitrates and nitrites with the participation of NADH₂, NAD(P)H or reduced quinone to ammonium (Lengeler et al., 2005; Kozlova et al., 2008). NarGHI nitrate reductase is an enzyme complex consisting of multi-heme b-type cytochrome, proteins with Fe-S clusters and Mo-containing cofactor (Morozkina & Zvyagilskaya, 2007; Kozlova et al., 2008). Nitrate reduction with the formation of nitrites and their subsequent reduction by a complex of periplasmic dissimilatory nitrite reductases to NH₄⁺ in *Wolinella succinogenes* and *Desulfuromonas* sp. was described (Bokranz et al., 1983; Chayka & Peretyatko, 2018).

Technogenically altered ecotopes, in particular such as Yavorivskoe Lake, are characterized by complex pollution, so several electron acceptors are available to bacteria at the same time. Strains of microorganisms isolated from such ecotopes are adapted to unfavourable conditions and therefore have high biotechnological potential. The selection of strains isolated from polluted ecotopes and adapted to contaminations which are capable of metabolizing a wide spectrum of pollutants is a particularly relevant task for establishing the mechanisms of their resistance and creation of new methods for environment purification (Mustapha & Halimoon, 2015). Studies of the influence of hexavalent chromium on sulfur,

nitrate and nitrite ions reduction by bacteria of *Desulfuromonas* sp. are important for development of effective and profitable biological methods of regulating their level in transformed biotopes. Investigation of the ability of sulfur reducing bacteria to reduce Cr(VI) and other electron acceptors is necessary for deepening understanding of mechanisms of their resistance and adaptation to existence in environments contaminated by chromium compounds. Therefore, the purpose of this work was to investigate the regularities of elemental sulfur, nitrate or nitrite ions' usage by sulfidogenic bacteria of *Desulfuromonas* genus at conditions of simultaneous presence in the media of another electron acceptor – Cr(VI), to establish the succession of electron acceptors' reduction by strains of these bacteria which were isolated by us from Yavorivske Lake and to evaluate the efficiency of their possible application in technologies of complex purification of the environment from metal compounds and other inorganic toxicants.

Materials and methods

Sulfur reducing bacteria *Desulfuromonas acetoxidans* IMV B-7384, *Desulfuromonas* sp. Yavor-5 and *Desulfuromonas* sp. Yavor-7, isolated by us earlier from Yavorivske Lake, were identified at the Microbiology Department of Ivan Franko National University of Lviv (Moroz et al., 2013). Strain *D. acetoxidans* IMV B-7384 has been stored in the depository of D. K. Zabolotny Institute of Microbiology and Virology of the NAS of Ukraine since 2013.

The bacteria were grown in Kravtsov-Sorokin media (Gudz et al., 2014) without SO_4^{2-} and without Mohr's salt of such composition (g/L): $\text{NaH}_2\text{PO}_4 \times 12\text{H}_2\text{O}$ (0.84), K_2HPO_4 (0.5), NH_4Cl (0.16), $\text{MgCl}_2 \times 6\text{H}_2\text{O}$ (0.10), sodium lactate ($\text{NaC}_3\text{H}_5\text{O}_3$) (2.00). Before bacteria seeding 0.05 mL of $\text{Na}_2\text{S} \times 9\text{H}_2\text{O}$ (1%) sterile solution was added to the media. A sterile 10 N NaOH solution was used to provide pH of the media to 7.2. Bacteria were sown in the media to initial concentration of cells of 0.1 mg/mL. Solutions of sodium fumarate ($\text{C}_4\text{H}_3\text{NaO}_4$), NaNO_3 , NaNO_2 , $\text{K}_2\text{Cr}_2\text{O}_7$ were sterilized separately and placed into the media before seeding of the cells at different concentrations. S^0 was sterilized separately (0.5 atm) and placed in the media as weighted quantities (0.11 g/L) at concentration of 3.47 mM (concentration of SO_4^{2-} in media of standard composition). Into the media with $\text{C}_4\text{H}_3\text{NaO}_4$, $\text{K}_2\text{Cr}_2\text{O}_7$ or NaNO_3 or NaNO_2 0.017 mM cysteine ($\text{C}_3\text{H}_7\text{NO}_2\text{S}$) was introduced to provide the assimilation needs of bacteria in sulfur (Lengeler et al., 2005). To media with NaNO_3 , NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ or without it NH_4Cl was not added. Bacteria were grown for 10 days in test tubes (25 mL), completely topped up by the media and tightly closed with rubber plugs, at a temperature of 30 °C.

To determine the efficiency of sulfur, nitrate, or nitrite ions' reduction at simultaneous presence in the media of potassium dichromate (the media with two electron acceptors: S^0 , NO_3^- or NO_2^- and Cr(VI)), cells were previously cultivated in the media with sodium fumarate (3.47 mM) as an electron acceptor and sodium lactate (17.86 mM) as an electron donor to the middle of the exponential growth phase. Bacteria were sown in a media with sodium lactate (17.86 mM), to which weighted quantities of insoluble in water sterile S^0 and sterile 1 M solutions of NaNO_3 or NaNO_2 were added to their final concentration in the media of 3.47 mM (concentration of SO_4^{2-} in media of standard composition) and different volumes of the sterile 1 M solution of $\text{K}_2\text{Cr}_2\text{O}_7$ to final Cr(VI) concentrations in the media of 1.74, 3.47, 5.21, 6.94 and 10.41 mM, which is in 0.5, 1.0, 1.5, 2.0 and 3.0 times different from the standard electron acceptor content in Kravtsov-Sorokin media. Bacteria were also sown in a media with sodium lactate at the same concentration, to which different volumes of sterile 1 M solutions of NaNO_3 or NaNO_2 were added to their final concentrations in the media of 1.74, 3.47, 5.21, 6.94 and 10.41 mM and the sterile 1 M solution of $\text{K}_2\text{Cr}_2\text{O}_7$ to final Cr(VI) concentration in the media of 3.47 mM. The cells were also sown in a media with sodium lactate, to which were added weighted quantities of S^0 , sterile 1 M solutions of NaNO_3 , NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final S^0 , NO_3^- , NO_2^- or Cr(VI) concentration in the media of 3.47 mM, to test the bacteria growth in media with sulfur, nitrate, nitrite ions or Cr(VI) as the sole electron acceptor (control). Into the media without bacteria the weighted quantities of S^0 , solutions of NaNO_3 , NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ were added to final S^0 , NO_3^- , NO_2^- or Cr(VI) concentration in the media of 3.47 mM to verify their spontaneous reduction.

Different volumes of the 1 M solution of $\text{K}_2\text{Cr}_2\text{O}_7$ in the media to obtain required Cr(VI) concentrations were calculated taking into account the molecular weight of the compound and atomic mass of chromium (two atoms), since it is known that in aqueous solutions dichromate ions are hydrolysed to form of HCrO_4^- : $\text{Cr}_2\text{O}_7^{2-} + \text{H}_2\text{O} \rightleftharpoons 2\text{HCrO}_4^-$ (Mandich, 1997). Therefore, to obtain the same content of S^0 , NO_3^- , NO_2^- or Cr(VI) the molar concentration of $\text{K}_2\text{Cr}_2\text{O}_7$ in the media was twice lower than S^0 and compounds of nitrogen (NaNO_3 or NaNO_2) that contain one atom of sulfur or nitrogen. Biomass, the concentrations of nitrate or nitrite ions, Cr(VI), Cr(III), hydrogen sulfide or ammonium ions in cultural liquid were determined on 10 day or 1, 2, 4, 6, 8 and 10 days of growth. By the difference between the initial and residual content of electron acceptors in the media the efficiency (%) of their reduction by bacteria was calculated, based on the ratio of molar concentrations of reduced by bacteria nitrate, nitrite ions or Cr(VI) in the process of anaerobic respiration and their concentrations at the beginning of cultivation, which were taken as 100%.

Biomass was determined by the turbidimetric method by the optical density of the cell suspension by measuring it at a wavelength of 340 nm in a cuvette with an optical way of 3 mm and calculated using the formula: $C, \text{ g/L} = (E_{340} \times n)/K$, where E_{340} – extinction ($\lambda = 340 \text{ nm}$); n – dilution factor; K – coefficient of recalculation, obtained from the calibration curve of the dependence of extinction from the mass of dry cells, determined by the weight method, and equal to 0.72 (Gudz et al., 2014). In a cultural liquid, separated from the cells by centrifugation (4025 g, 15 min), the concentrations of nitrate ions (after their reduction to nitrites in the presence of Zn:MnSO₄ (1:100) powder as a reducing agent) and nitrite ions were determined by spectrophotometric method which relies on a diazotization reaction with a Griess reagent (n-(1-naphthyl)ethylenediamine dihydrochloride, sulfanil and acetic acid) (Granger, 1996), Cr(VI) colorimetrically by interaction with 1,5-diphenylcarbazide in acid solution, Cr(III) by reaction with chromazurol S, hydrogen sulfide by spectrophotometric method for the formation of methylene blue and ammonium ions by colorimetric method for the formation of indophenol (Gudz et al., 2014).

Experiments were repeated three times with three parallel formulations for each variant of experimental and control conditions. The obtained data were processed by generally accepted methods of variation statistics. The data in the tables are presented as $x \pm \text{SD}$ (mean value \pm standard deviation of three measurements). The reliability of the difference between experimental and control variants was evaluated using ANOVA software. Differences between the samples were considered statistically significant at $P < 0.05$.

Results

The intensity of anaerobic respiration of microorganisms in contaminated ecotopes is determined by the level of their adaptation to unfavourable environmental conditions. The efficiency of technologies of complex purification of the environment from pollutants primarily depends from resistance of the selected strains of bacteria to metal compounds, in particular, increased content of hexavalent chromium. Therefore, we studied the ability of bacteria *Desulfuromonas* sp. to reduce in the process of anaerobic respiration elemental sulfur, nitrate or nitrite ions with the simultaneous presence in the media of $\text{K}_2\text{Cr}_2\text{O}_7$ at different concentrations.

To study the influence of sodium dichromate at Cr(VI) concentration in the media of 1.74–10.41 mM on the sulfur reduction by sulfur reducing bacteria, they were grown in the media, to which 3.47 mM S^0 and $\text{K}_2\text{Cr}_2\text{O}_7$ at different concentrations were added. The bacteria were also sown in the media with S^0 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final sulfur or Cr(VI) concentration in the media of 3.47 mM, to test the use by bacteria of sulfur or hexavalent chromium as the sole electron acceptor (Table 1). After 10 days of growth the biomass of bacteria in the media with S^0 was 1.7–2.0 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$. After the simultaneous addition of S^0 and $\text{K}_2\text{Cr}_2\text{O}_7$ to the cultivation media with growing of Cr(VI) concentrations a gradual decrease in the biomass accumulation by bacteria was observed, compared to growth in the media with only S^0 . In the media with 3.47 mM S^0 and 10.41 mM Cr(VI) the growth of bacteria decreased 2.3–2.4 times, compared with growth in the media with only S^0 . In the media with S^0 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increase of Cr(VI) concentrations a

gradual decrease was observed in concentrations of hydrogen sulfide produced by bacteria, as compared with its production in the media with only S^0 . In media with S^0 and $K_2Cr_2O_7$ the cells produced 0.49–0.74 mM hydrogen sulfide (control: 2.36–2.40 mM, Table 1). The efficiency of Cr(VI) reduction by bacteria in the media with S^0 and $K_2Cr_2O_7$ was found to be 1.1–2.0 times lower as compared with its reduction in the media with only $K_2Cr_2O_7$ (70.6–72.1%, Fig. 1). In the media with sulfur and $K_2Cr_2O_7$ bacteria produced 1.08–3.88 mM of Cr(III) (control: 2.43–2.47 mM, Table 1). In the media with S^0 or $K_2Cr_2O_7$ without bacteria the efficiency of spontaneous sulfur and Cr(VI) reduction was found to be insignificant and did not exceed 4.0 (calculated according to the produced H_2S) and 4.3% respectively (Table 1, Fig. 1). Thus, it has been established that Cr(VI) inhibits the biomass accumulation and hydrogen sulfide production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 3.47 mM S^0 and 1.74–10.41 mM Cr(VI). In the media with the same initial content (3.47 mM) of S^0 and Cr(VI) bacteria produced Cr(III) at concentrations 3.3–3.4 times higher than that of hydrogen sulfide.

For research on the influence of sodium dichromate at Cr(VI) concentration in the media of 1.74–10.41 mM on the 3.47 mM nitrate ions' usage by bacteria, they were cultivated in the media without NH_4Cl , to which 3.47 mM $NaNO_3$ and $K_2Cr_2O_7$ at different concentrations were added. The bacteria were also grown in the media with $NaNO_3$ or $K_2Cr_2O_7$ to final NO_3^- or Cr(VI) concentration in the media of 3.47 mM, to test the usage by bacteria of nitrate ions or hexavalent chromium as the sole electron acceptor (Table 2). After 10 days of growth the biomass of bacteria in the media with $NaNO_3$ was revealed to be 2.0–2.1 times higher than in the media with $K_2Cr_2O_7$. After simultaneous addition into the media of $NaNO_3$ and $K_2Cr_2O_7$ with increasing concentrations of Cr(VI) a gradual inhibition of bacteria growth was observed, compared with growth in the media with only $NaNO_3$. In the media with NO_3^- and 10.41 mM Cr(VI) the growth of bacteria decreased 2.2–2.3 times, compared with the growth in the media with $NaNO_3$ as the sole electron acceptor. In the media with $NaNO_3$ and $K_2Cr_2O_7$ with increase of Cr(VI) concentrations a gradual (1.2–2.2 times) decrease was also observed in the efficiency of nitrate ions' reduction by bacteria, compared with their reduction in the media with only $NaNO_3$ (92.7–94.8%) (Fig. 2a). In the media with $NaNO_3$ and $K_2Cr_2O_7$ bacteria produced 1.48–2.58 mM of ammonium ions (control: 2.69–2.74 mM, Table 2). The efficiency of

Cr(VI) reduction by cells with increase in its concentrations in the media with $NaNO_3$ and $K_2Cr_2O_7$ was revealed to be from 1.2 to 2.3 times lower than its reduction in the media only with $K_2Cr_2O_7$ (69.5–70.3%, Fig. 2b). In the media with $NaNO_3$ and $K_2Cr_2O_7$ bacteria produced 0.94–3.34 mM of Cr(III) (control: 2.38–2.39 mM, Table 2). In a media with $NaNO_3$ or $K_2Cr_2O_7$ without bacteria the efficiency of NO_3^- and Cr(VI) reduction did not exceed 3.7% and 4.0%, respectively (Fig. 2). Thus, it has been shown that $K_2Cr_2O_7$ inhibits biomass accumulation, nitrate ions' reduction and ammonium ions' production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 3.47 mM NO_3^- and Cr(VI) (1.74–10.41 mM).

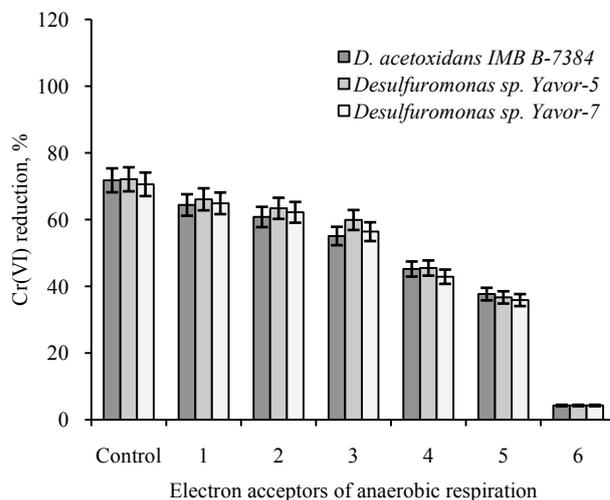


Fig. 1. Efficiency of 1.74–10.41 mM Cr(VI) reduction by *Desulfuromonas* sp. after 10 days of growth in media with S^0 or $K_2Cr_2O_7$ ($x \pm SD$, $n = 3$): designation on the horizontal axis: control – 3.47 mM Cr(VI); 1 – 3.47 mM S^0 and 1.74 mM Cr(VI); 2 – 3.47 mM S^0 and 3.47 mM Cr(VI); 3 – 3.47 mM S^0 and 5.21 mM Cr(VI); 4 – 3.47 mM S^0 and 6.94 mM Cr(VI); 5 – 3.47 mM S^0 and 10.41 mM Cr(VI); 6 – 3.47 mM Cr(VI) (without bacteria)

Table 1

Reduction of 3.47 mM S^0 and 1.74–10.41 mM Cr(VI) by *Desulfuromonas* sp. after 10 days of growth in media with S^0 or $K_2Cr_2O_7$ ($x \pm SD$, $n = 3$)

Strain	Electron acceptors of anaerobic respiration	Residual content of Cr(VI) in cultural liquid, mM	Cr(III), mM	S^{2-} , mM	Biomass, g/L
<i>D. acetoxidans</i> IMB B-7384	3.47 mM S^0	0	0	2.36 ± 0.02	2.17 ± 0.02
	3.47 mM S^0 (wb)	0	0	0.14 ± 0.02	0
	3.47 mM S^0 and 1.74 mM Cr(VI)	0.62 ± 0.09	1.08 ± 0.01	0.71 ± 0.04	1.88 ± 0.07
	3.47 mM S^0 and 3.47 mM Cr(VI)	1.36 ± 0.01	2.08 ± 0.02	0.62 ± 0.07	1.69 ± 0.08
	3.47 mM S^0 and 5.21 mM Cr(VI)	2.34 ± 0.08	2.76 ± 0.02	0.59 ± 0.02	1.30 ± 0.04
	3.47 mM S^0 and 6.94 mM Cr(VI)	3.80 ± 0.09	3.07 ± 0.06	0.53 ± 0.03	1.15 ± 0.09
	3.47 mM S^0 and 10.41 mM Cr(VI)	6.49 ± 0.01	3.88 ± 0.04	0.51 ± 0.07	0.93 ± 0.01
	3.47 mM Cr(VI)	0.98 ± 0.08	2.45 ± 0.04	0	1.25 ± 0.02
<i>Desulfuromonas</i> sp. Yavor-5	3.47 mM Cr(VI) (wb)	3.32 ± 0.05	0.15 ± 0.01	0	0
	3.47 mM S^0	0	0	2.40 ± 0.02	2.23 ± 0.08
	3.47 mM S^0 (wb)	0	0	0.14 ± 0.02	0
	3.47 mM S^0 and 1.74 mM Cr(VI)	0.59 ± 0.09	1.10 ± 0.06	0.73 ± 0.04	1.80 ± 0.01
	3.47 mM S^0 and 3.47 mM Cr(VI)	1.27 ± 0.07	2.11 ± 0.05	0.65 ± 0.07	1.61 ± 0.08
	3.47 mM S^0 and 5.21 mM Cr(VI)	2.09 ± 0.06	3.02 ± 0.07	0.58 ± 0.06	1.26 ± 0.07
	3.47 mM S^0 and 6.94 mM Cr(VI)	3.78 ± 0.02	3.12 ± 0.03	0.51 ± 0.03	1.13 ± 0.06
	3.47 mM S^0 and 10.41 mM Cr(VI)	6.59 ± 0.01	3.79 ± 0.07	0.49 ± 0.03	0.98 ± 0.06
<i>Desulfuromonas</i> sp. Yavor-7	3.47 mM Cr(VI)	0.97 ± 0.01	2.47 ± 0.03	0	1.20 ± 0.06
	3.47 mM Cr(VI) (wb)	3.32 ± 0.05	0.15 ± 0.01	0	0
	3.47 mM S^0	0	0	2.38 ± 0.01	2.32 ± 0.03
	3.47 mM S^0 (wb)	0	0	0.14 ± 0.02	0
	3.47 mM S^0 and 1.74 mM Cr(VI)	0.61 ± 0.01	1.09 ± 0.06	0.74 ± 0.04	1.92 ± 0.01
	3.47 mM S^0 and 3.47 mM Cr(VI)	1.31 ± 0.05	2.10 ± 0.05	0.63 ± 0.07	1.67 ± 0.08
	3.47 mM S^0 and 5.21 mM Cr(VI)	2.27 ± 0.04	2.89 ± 0.07	0.56 ± 0.06	1.41 ± 0.07
	3.47 mM S^0 and 6.94 mM Cr(VI)	3.96 ± 0.06	2.95 ± 0.03	0.54 ± 0.03	1.22 ± 0.06
3.47 mM S^0 and 10.41 mM Cr(VI)	6.67 ± 0.01	3.69 ± 0.07	0.50 ± 0.04	0.96 ± 0.06	
3.47 mM Cr(VI)	1.02 ± 0.02	2.43 ± 0.02	0	1.17 ± 0.06	
3.47 mM Cr(VI) (wb)	3.32 ± 0.05	0.15 ± 0.01	0	0	

Note: (wb) – the media without bacteria.

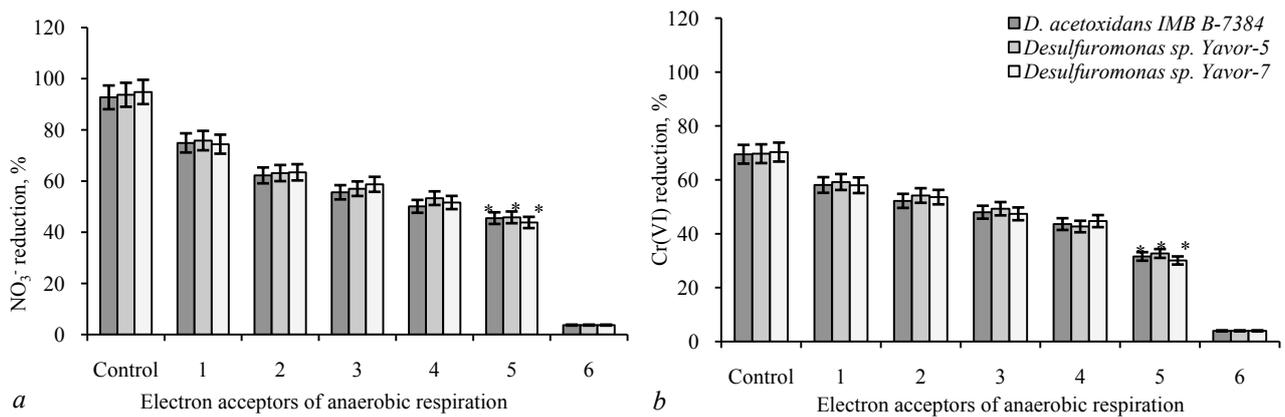


Fig. 2. Efficiency of 3.47 mM NO_3^- (a) and 1.74–10.41 mM Cr(VI) (b) reduction by *Desulfuromonas* sp. after 10 days of growth in media with NaNO_3 and/or $\text{K}_2\text{Cr}_2\text{O}_7$ ($x \pm \text{SD}$, $n = 3$): designation on the horizontal axis: control – 3.47 mM nitrate ions (a), 3.47 mM Cr(VI) (b); 1 – 3.47 mM nitrate ions and 1.74 mM Cr(VI); 2 – 3.47 mM nitrate ions and 3.47 mM Cr(VI); 3 – 3.47 mM nitrate ions and 5.21 mM Cr(VI); 4 – 3.47 mM nitrate ions and 6.94 mM Cr(VI); 5 – 3.47 mM nitrate ions and 10.41 mM Cr(VI); 6 – 3.47 mM nitrate ions (without bacteria) (a), 3.47 mM Cr(VI) (without bacteria) (b); * – the data were statistically significant as compared with the control ($P < 0.05$)

Table 2

Reduction of 3.47 mM NO_3^- and 1.74–10.41 mM Cr(VI) by *Desulfuromonas* sp. after 10 days of growth in media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ ($x \pm \text{SD}$, $n = 3$)

Strain	Electron acceptors of anaerobic respiration	Residual content in cultural liquid, mM		Cr(III), mM	NH_4^+ , mM	Biomass, g/L
		NO_3^-	Cr(VI)			
<i>D. acetoxidans</i> IMB B-7384	3.47 mM NO_3^-	0.25 ± 0.02	0	0	2.69 ± 0.02	2.47 ± 0.02
	3.47 mM NO_3^- (wb)	3.34 ± 0.03	0	0	0.11 ± 0.01	0
	3.47 mM NO_3^- and 1.74 mM Cr(VI)	0.87 ± 0.01	0.73 ± 0.03	0.96 ± 0.04	2.53 ± 0.05	2.22 ± 0.03
	3.47 mM NO_3^- and 3.47 mM Cr(VI)	1.31 ± 0.02	1.66 ± 0.02	1.76 ± 0.08	2.11 ± 0.01	1.76 ± 0.05
	3.47 mM NO_3^- and 5.21 mM Cr(VI)	1.54 ± 0.02	2.71 ± 0.05	2.46 ± 0.01	1.91 ± 0.02	1.54 ± 0.01
	3.47 mM NO_3^- and 6.94 mM Cr(VI)	1.73 ± 0.04	3.91 ± 0.08	3.01 ± 0.02	1.69 ± 0.01	1.31 ± 0.01
	3.47 mM NO_3^- and 10.41 mM Cr(VI)	1.89 ± 0.04	7.12 ± 0.01	3.24 ± 0.03	1.53 ± 0.03	1.09 ± 0.02
	3.47 mM Cr(VI)	0	1.06 ± 0.02	2.38 ± 0.04	0	1.25 ± 0.05
	3.47 mM Cr(VI)(wb)	0	3.33 ± 0.06	0.13 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-5	3.47 mM NO_3^-	0.22 ± 0.01	0	0	2.73 ± 0.06	2.45 ± 0.04
	3.47 mM NO_3^- (wb)	3.34 ± 0.03	0	0	0.11 ± 0.01	0
	3.47 mM NO_3^- and 1.74 mM Cr(VI)	0.84 ± 0.03	0.71 ± 0.08	0.97 ± 0.01	2.58 ± 0.08	2.19 ± 0.07
	3.47 mM NO_3^- and 3.47 mM Cr(VI)	1.28 ± 0.08	1.59 ± 0.02	1.84 ± 0.08	2.15 ± 0.09	1.86 ± 0.08
	3.47 mM NO_3^- and 5.21 mM Cr(VI)	1.49 ± 0.05	2.64 ± 0.03	2.55 ± 0.02	1.94 ± 0.04	1.63 ± 0.01
	3.47 mM NO_3^- and 6.94 mM Cr(VI)	1.62 ± 0.02	3.98 ± 0.06	2.93 ± 0.04	1.79 ± 0.02	1.25 ± 0.03
	3.47 mM NO_3^- and 10.41 mM Cr(VI)	1.88 ± 0.04	7.01 ± 0.08	3.34 ± 0.09	1.55 ± 0.03	1.10 ± 0.02
	3.47 mM Cr(VI)	0	1.05 ± 0.04	2.38 ± 0.02	0	1.19 ± 0.04
	3.47 mM Cr(VI)(wb)	0	3.33 ± 0.06	0.13 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-7	3.47 mM NO_3^-	0.18 ± 0.01	0	0	2.74 ± 0.02	2.51 ± 0.03
	3.47 mM NO_3^- (wb)	3.34 ± 0.03	0	0	0.11 ± 0.01	0
	3.47 mM NO_3^- and 1.74 mM Cr(VI)	0.89 ± 0.05	0.73 ± 0.08	0.94 ± 0.02	2.55 ± 0.06	2.23 ± 0.01
	3.47 mM NO_3^- and 3.47 mM Cr(VI)	1.27 ± 0.03	1.61 ± 0.02	1.81 ± 0.03	2.17 ± 0.09	1.94 ± 0.03
	3.47 mM NO_3^- and 5.21 mM Cr(VI)	1.43 ± 0.04	2.74 ± 0.03	2.43 ± 0.04	2.01 ± 0.04	1.71 ± 0.05
	3.47 mM NO_3^- and 6.94 mM Cr(VI)	1.68 ± 0.02	3.84 ± 0.06	3.08 ± 0.03	1.75 ± 0.05	1.39 ± 0.08
	3.47 mM NO_3^- and 10.41 mM Cr(VI)	1.95 ± 0.06	7.28 ± 0.08	3.11 ± 0.07	1.48 ± 0.04	1.12 ± 0.01
	3.47 mM Cr(VI)	0	1.03 ± 0.04	2.39 ± 0.06	0	1.21 ± 0.02
	3.47 mM Cr(VI)(wb)	0	3.33 ± 0.06	0.13 ± 0.01	0	0

Notes: (wb) – the media without bacteria; to media with NO_3^- and Cr(VI) or without it NH_4Cl was not added.

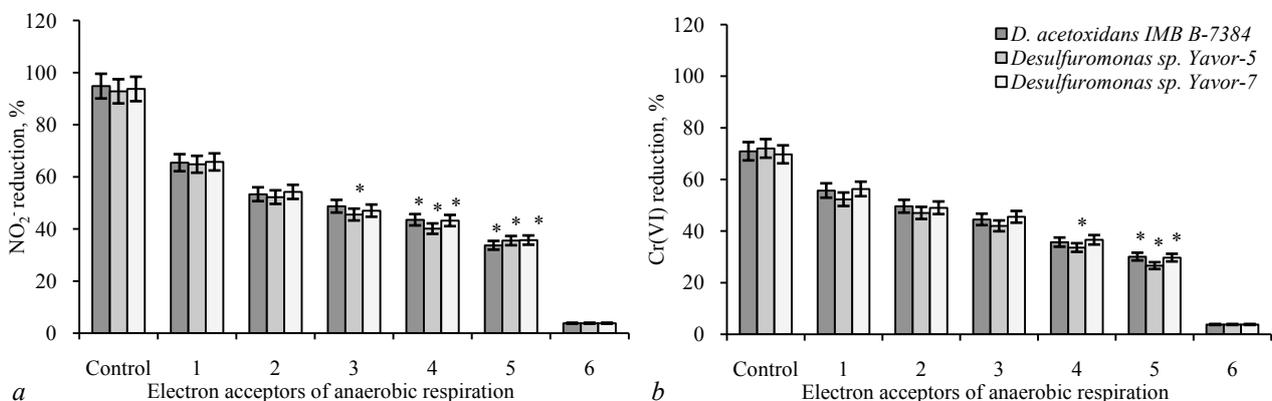


Fig. 3. Efficiency of 3.47 mM NO_2^- (a) and 1.74–10.41 mM Cr(VI) (b) reduction by *Desulfuromonas* sp. after 10 days of growth in media with NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ ($x \pm \text{SD}$, $n = 3$): designation on the horizontal axis: control – 3.47 mM nitrite ions (a), 3.47 mM Cr(VI) (b); 1 – 3.47 mM nitrite ions and 1.74 mM Cr(VI); 2 – 3.47 mM nitrite ions and 3.47 mM Cr(VI); 3 – 3.47 mM nitrite ions and 5.21 mM Cr(VI); 4 – 3.47 mM nitrite ions and 6.94 mM Cr(VI); 5 – 3.47 mM nitrite ions and 10.41 mM Cr(VI); 6 – 3.47 mM nitrite ions (without bacteria) (a), 3.47 mM Cr(VI) (without bacteria) (b); * – the data were statistically significant as compared with the control ($P < 0.05$)

Table 3Reduction of 3.47 mM NO₂⁻ and 1.74–10.41 mM Cr(VI) by *Desulfuromonas* sp. after 10 days of growth in media with NaNO₂ or K₂Cr₂O₇ (x ± SD, n = 3)

Strain	Electron acceptors of anaerobic respiration	Residual content in cultural liquid, mM		Cr(III), mM	NH ₄ ⁺ , mM	Biomass, g/L
		NO ₂ ⁻	Cr(VI)			
<i>D. acetoxidans</i> IMV B-7384	3.47 mM NO ₂ ⁻	0.18 ± 0.01	0	0	2.59 ± 0.02	2.36 ± 0.06
	3.47 mM NO ₂ ⁻ (wb)	3.34 ± 0.02	0	0	0.09 ± 0.01	0
	3.47 mM NO ₂ ⁻ and 1.74 mM Cr(VI)	1.20 ± 0.05	0.77 ± 0.03	0.95 ± 0.02	2.25 ± 0.02	2.12 ± 0.01
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.62 ± 0.03	1.75 ± 0.02	1.71 ± 0.03	1.80 ± 0.05	1.64 ± 0.03
	3.47 mM NO ₂ ⁻ and 5.21 mM Cr(VI)	1.78 ± 0.04	2.89 ± 0.03	2.30 ± 0.02	1.64 ± 0.04	1.41 ± 0.05
	3.47 mM NO ₂ ⁻ and 6.94 mM Cr(VI)	1.96 ± 0.02	4.46 ± 0.06	2.46 ± 0.02	1.46 ± 0.05	1.29 ± 0.08
	3.47 mM NO ₂ ⁻ and 10.41 mM Cr(VI)	2.30 ± 0.06	7.28 ± 0.08	3.09 ± 0.07	1.15 ± 0.02	1.02 ± 0.01
	3.47 mM Cr(VI)	0	1.01 ± 0.04	2.41 ± 0.02	0	1.17 ± 0.02
<i>Desulfuromonas</i> sp. Yavor-5	3.47 mM NO ₂ ⁻	0.25 ± 0.06	0	0	2.55 ± 0.02	2.32 ± 0.01
	3.47 mM NO ₂ ⁻ (wb)	3.34 ± 0.02	0	0	0.09 ± 0.01	0
	3.47 mM NO ₂ ⁻ and 1.74 mM Cr(VI)	1.22 ± 0.01	0.83 ± 0.03	0.89 ± 0.03	2.22 ± 0.03	2.11 ± 0.03
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.66 ± 0.02	1.84 ± 0.02	1.60 ± 0.03	1.78 ± 0.03	1.66 ± 0.05
	3.47 mM NO ₂ ⁻ and 5.21 mM Cr(VI)	1.89 ± 0.02	3.02 ± 0.05	2.15 ± 0.01	1.54 ± 0.04	1.44 ± 0.01
	3.47 mM NO ₂ ⁻ and 6.94 mM Cr(VI)	2.08 ± 0.06	4.61 ± 0.08	2.31 ± 0.02	1.35 ± 0.01	1.21 ± 0.01
	3.47 mM NO ₂ ⁻ and 10.41 mM Cr(VI)	2.24 ± 0.04	7.64 ± 0.01	2.73 ± 0.03	1.19 ± 0.03	0.99 ± 0.02
	3.47 mM Cr(VI)	0	0.97 ± 0.02	2.42 ± 0.04	0	1.25 ± 0.05
<i>Desulfuromonas</i> sp. Yavor-7	3.47 mM NO ₂ ⁻	0.22 ± 0.01	0	0	2.53 ± 0.06	2.40 ± 0.03
	3.47 mM NO ₂ ⁻ (wb)	3.34 ± 0.02	0	0	0.09 ± 0.01	0
	3.47 mM NO ₂ ⁻ and 1.74 mM Cr(VI)	1.19 ± 0.03	0.76 ± 0.02	0.96 ± 0.02	2.24 ± 0.04	2.14 ± 0.07
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.59 ± 0.02	1.77 ± 0.02	1.69 ± 0.01	1.82 ± 0.06	1.66 ± 0.08
	3.47 mM NO ₂ ⁻ and 5.21 mM Cr(VI)	1.84 ± 0.05	2.84 ± 0.03	2.33 ± 0.04	1.59 ± 0.04	1.43 ± 0.01
	3.47 mM NO ₂ ⁻ and 6.94 mM Cr(VI)	1.97 ± 0.02	4.40 ± 0.06	2.52 ± 0.04	1.48 ± 0.02	1.25 ± 0.03
	3.47 mM NO ₂ ⁻ and 10.41 mM Cr(VI)	2.23 ± 0.04	7.32 ± 0.03	3.05 ± 0.04	1.22 ± 0.03	1.10 ± 0.01
	3.47 mM Cr(VI)	0	1.05 ± 0.04	2.37 ± 0.03	0	1.19 ± 0.04
3.47 mM Cr(VI) (wb)	0	3.34 ± 0.05	0.12 ± 0.02	0	0	

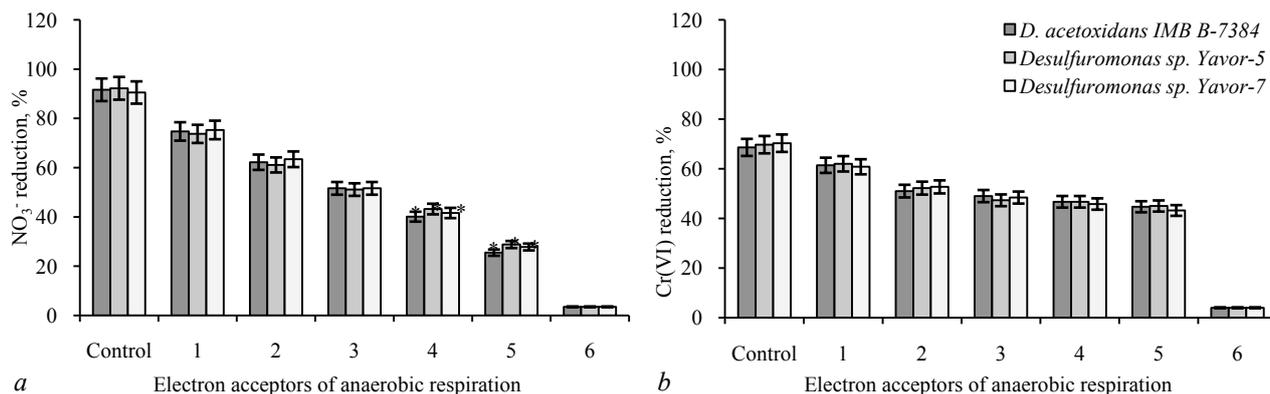
Notes: (wb) – the media without bacteria; to media with NO₂⁻ and Cr(VI) or without it the NH₄Cl was not added.

Fig. 4. Efficiency of 1.74–10.41 mM NO₃⁻ (a) and 3.47 mM Cr(VI) (b) reduction by *Desulfuromonas* sp. after 10 days of growth in media with NaNO₃ or K₂Cr₂O₇ (x ± SD, n = 3): designation on the horizontal axis: control – 3.47 mM nitrate ions (a), 3.47 mM Cr(VI) (b); 1 – 1.74 mM nitrate ions and 3.47 mM Cr(VI); 2 – 3.47 mM nitrate ions and 3.47 mM Cr(VI); 3 – 5.21 mM nitrate ions and 3.47 mM Cr(VI); 4 – 6.94 mM nitrate ions and 3.47 mM Cr(VI); 5 – 10.41 mM nitrate ions and 3.47 mM Cr(VI); 6 – 3.47 mM nitrate ions (without bacteria) (a), 3.47 mM Cr(VI) (without bacteria) (b); * – the data were statistically significant as compared with the control (P < 0.05)

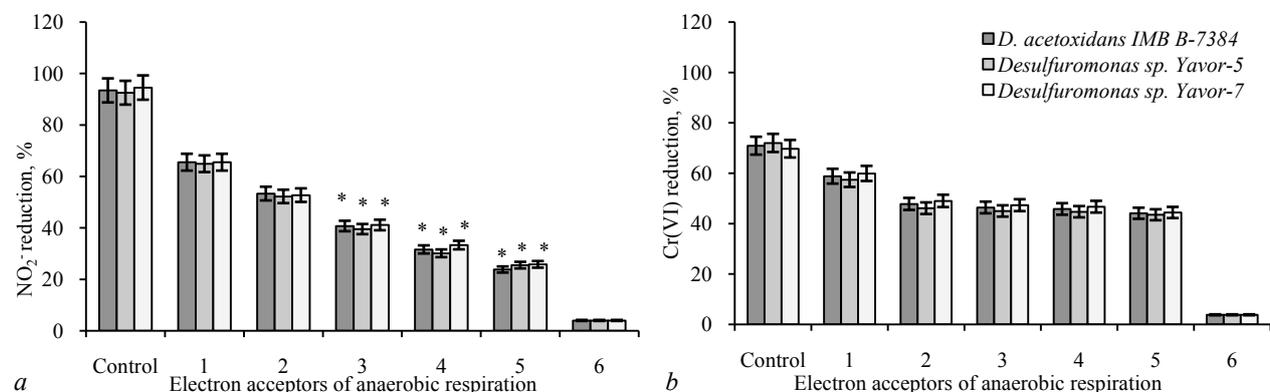


Fig. 5. Efficiency of 1.74–10.41 mM NO₂⁻ (a) and 3.47 mM Cr(VI) (b) reduction by *Desulfuromonas* sp. after 10 days of growth in media with NaNO₂ or K₂Cr₂O₇ (x ± SD, n = 3): designation on the horizontal axis: control – 3.47 mM nitrite ions (a), 3.47 mM Cr(VI) (b); 1 – 1.74 mM nitrite ions and 3.47 mM Cr(VI); 2 – 3.47 mM nitrite ions and 3.47 mM Cr(VI); 3 – 5.21 mM nitrite ions and 3.47 mM Cr(VI); 4 – 6.94 mM nitrite ions and 3.47 mM Cr(VI); 5 – 10.41 mM nitrite ions and 3.47 mM Cr(VI); 6 – 3.47 mM nitrite ions (without bacteria) (a), 3.47 mM Cr(VI) (without bacteria) (b); * – the data were statistically significant as compared with the control (P < 0.05)

Table 4Reduction of 1.74–10.41 mM NO₃⁻ and 3.47 mM Cr(VI) by *Desulfuromonas* sp. after 10 days of growth in media with NaNO₃ or K₂Cr₂O₇ (x ± SD, n = 3)

Strain	Electron acceptors of anaerobic respiration	Residual content in cultural liquid, mM		Cr(III), mM	NH ₄ ⁺ , mM	Biomass, g/L
		NO ₃ ⁻	Cr(VI)			
<i>D. acetoxidans</i> IMV B-7384	3.47 mM NO ₃ ⁻	0.29 ± 0.04	0	0	2.69 ± 0.02	2.49 ± 0.01
	3.47 mM NO ₃ ⁻ (wb)	3.35 ± 0.03	0	0	0.09 ± 0.01	0
	1.74 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	0.44 ± 0.01	1.34 ± 0.03	2.11 ± 0.02	1.27 ± 0.03	2.22 ± 0.03
	3.47 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	1.31 ± 0.02	1.70 ± 0.02	1.75 ± 0.02	2.14 ± 0.01	1.73 ± 0.03
	5.21 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	2.52 ± 0.02	1.77 ± 0.05	1.70 ± 0.01	2.65 ± 0.04	1.51 ± 0.05
	6.94 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	4.16 ± 0.06	1.85 ± 0.05	1.62 ± 0.02	2.76 ± 0.02	1.21 ± 0.01
	10.41 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	7.76 ± 0.04	1.92 ± 0.01	1.53 ± 0.01	2.62 ± 0.03	0.98 ± 0.02
	3.47 mM Cr(VI)	0	1.09 ± 0.02	2.36 ± 0.02	0	1.25 ± 0.04
	3.47 mM Cr(VI) (wb)	0	3.33 ± 0.01	0.13 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-5	3.47 mM NO ₃ ⁻	0.27 ± 0.01	0	0	2.73 ± 0.06	2.46 ± 0.03
	3.47 mM NO ₃ ⁻ (wb)	3.35 ± 0.03	0	0	0.09 ± 0.01	0
	1.74 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	0.46 ± 0.03	1.32 ± 0.08	2.12 ± 0.03	1.26 ± 0.02	2.23 ± 0.07
	3.47 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	1.35 ± 0.04	1.66 ± 0.02	1.79 ± 0.01	2.10 ± 0.03	1.81 ± 0.05
	5.21 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	2.55 ± 0.05	1.83 ± 0.03	1.63 ± 0.04	2.63 ± 0.03	1.62 ± 0.04
	6.94 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	3.94 ± 0.02	1.85 ± 0.06	1.60 ± 0.04	2.96 ± 0.02	1.25 ± 0.03
	10.41 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	7.41 ± 0.04	1.91 ± 0.02	1.54 ± 0.01	2.95 ± 0.05	1.01 ± 0.01
	3.47 mM Cr(VI)	0	1.05 ± 0.04	2.42 ± 0.02	0	1.16 ± 0.03
	3.47 mM Cr(VI) (wb)	0	3.33 ± 0.01	0.13 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-7	3.47 mM NO ₃ ⁻	0.33 ± 0.01	0	0	2.70 ± 0.02	2.52 ± 0.06
	3.47 mM NO ₃ ⁻ (wb)	3.35 ± 0.03	0	0	0.09 ± 0.01	0
	1.74 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	0.43 ± 0.05	1.36 ± 0.08	2.07 ± 0.04	1.29 ± 0.02	2.26 ± 0.01
	3.47 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	1.27 ± 0.03	1.64 ± 0.02	1.81 ± 0.03	2.18 ± 0.02	1.74 ± 0.06
	5.21 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	2.52 ± 0.04	1.79 ± 0.03	1.67 ± 0.02	2.67 ± 0.04	1.54 ± 0.02
	6.94 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	4.05 ± 0.02	1.88 ± 0.06	1.57 ± 0.02	2.85 ± 0.04	1.29 ± 0.08
	10.41 mM NO ₃ ⁻ and 3.47 mM Cr(VI)	7.52 ± 0.06	1.97 ± 0.02	1.49 ± 0.03	2.88 ± 0.02	1.02 ± 0.01
	3.47 mM Cr(VI)	0	1.03 ± 0.04	2.43 ± 0.01	0	1.17 ± 0.02
	3.47 mM Cr(VI) (wb)	0	3.33 ± 0.01	0.13 ± 0.01	0	0

Notes: (wb) – the media without bacteria; to media with NO₃⁻ and Cr(VI) or without it NH₄Cl was not added.

In the media with the same initial content (3.47 mM) of NO₃⁻ and Cr(VI) bacteria reduced 1.2 times more nitrate ions than Cr(VI) with the production of ammonium ions at concentrations 1.2 times higher than that of Cr(III).

To investigate the influence of sodium dichromate at Cr(VI) concentration in the media of 1.74–10.41 mM on the 3.47 mM nitrite ions' reduction by sulfur reducing bacteria, they were grown in a media without NH₄Cl to which 3.47 mM NaNO₂ and K₂Cr₂O₇ at different concentra-

tions were added. The bacteria were also sown in a media with NaNO₂ or K₂Cr₂O₇ to final NO₂⁻ or Cr(VI) concentration in the media of 3.47 mM, to test the usage by bacteria of nitrite ions or hexavalent chromium as the sole electron acceptor (Table 3). Biomass of bacteria in the media with NaNO₂ was revealed to be up to 2.0 times higher than in the media with K₂Cr₂O₇. After simultaneous addition into the media of NaNO₂ and K₂Cr₂O₇ with increasing concentrations of Cr(VI) there was a decrease in the bacteria growth, compared with growth in the media with NaNO₂.

Table 5Reduction of 1.74–10.41 mM NO₂⁻ and 3.47 mM Cr(VI) by *Desulfuromonas* sp. after 10 days of growth in media with NaNO₂ or K₂Cr₂O₇ (x ± SD, n = 3)

Strain	Electron acceptors of anaerobic respiration	Residual content in cultural liquid, mM		Cr(III), mM	NH ₄ ⁺ , mM	Biomass, g/L
		NO ₂ ⁻	Cr(VI)			
<i>D. acetoxidans</i> IMV B-7384	3.47 mM NO ₂ ⁻	0.23 ± 0.01	0	0	2.56 ± 0.02	2.36 ± 0.05
	3.47 mM NO ₂ ⁻ (wb)	3.33 ± 0.02	0	0	0.12 ± 0.02	0
	1.74 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	0.60 ± 0.05	1.43 ± 0.08	2.04 ± 0.02	1.11 ± 0.06	2.12 ± 0.01
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.62 ± 0.03	1.81 ± 0.02	1.64 ± 0.02	1.82 ± 0.03	1.64 ± 0.03
	5.21 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	3.09 ± 0.04	1.86 ± 0.03	1.60 ± 0.02	2.09 ± 0.03	1.31 ± 0.05
	6.94 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	4.75 ± 0.02	1.88 ± 0.06	1.57 ± 0.03	2.17 ± 0.05	1.09 ± 0.08
	10.41 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	7.93 ± 0.06	1.94 ± 0.08	1.51 ± 0.03	2.44 ± 0.03	0.92 ± 0.01
	3.47 mM Cr(VI)	0	1.01 ± 0.04	2.45 ± 0.01	0	1.18 ± 0.04
	3.47 mM Cr(VI) (wb)	0	3.34 ± 0.05	0.12 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-5	3.47 mM NO ₂ ⁻	0.26 ± 0.06	0	0	2.59 ± 0.02	2.32 ± 0.01
	3.47 mM NO ₂ ⁻ (wb)	3.33 ± 0.02	0	0	0.12 ± 0.02	0
	1.74 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	0.61 ± 0.01	1.48 ± 0.03	1.97 ± 0.02	1.10 ± 0.05	2.15 ± 0.03
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.66 ± 0.02	1.87 ± 0.02	1.58 ± 0.04	1.79 ± 0.03	1.66 ± 0.05
	5.21 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	3.15 ± 0.02	1.91 ± 0.05	1.54 ± 0.01	2.03 ± 0.03	1.34 ± 0.01
	6.94 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	4.85 ± 0.06	1.92 ± 0.08	1.51 ± 0.03	2.03 ± 0.06	1.10 ± 0.01
	10.41 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	7.76 ± 0.04	1.96 ± 0.01	1.48 ± 0.03	2.62 ± 0.04	0.89 ± 0.02
	3.47 mM Cr(VI)	0	0.97 ± 0.02	2.48 ± 0.02	0	1.22 ± 0.03
	3.47 mM Cr(VI) (wb)	0	3.34 ± 0.05	0.12 ± 0.01	0	0
<i>Desulfuromonas</i> sp. Yavor-7	3.47 mM NO ₂ ⁻	0.19 ± 0.01	0	0	2.53 ± 0.06	2.30 ± 0.05
	3.47 mM NO ₂ ⁻ (wb)	3.33 ± 0.02	0	0	0.12 ± 0.02	0
	1.74 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	0.60 ± 0.03	1.39 ± 0.08	2.07 ± 0.01	1.13 ± 0.03	2.09 ± 0.07
	3.47 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	1.64 ± 0.05	1.77 ± 0.02	1.68 ± 0.03	1.81 ± 0.02	1.63 ± 0.05
	5.21 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	3.07 ± 0.05	1.83 ± 0.03	1.63 ± 0.01	2.10 ± 0.04	1.33 ± 0.04
	6.94 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	4.63 ± 0.02	1.85 ± 0.06	1.59 ± 0.04	2.28 ± 0.02	1.05 ± 0.03
	10.41 mM NO ₂ ⁻ and 3.47 mM Cr(VI)	7.72 ± 0.04	1.93 ± 0.08	1.54 ± 0.02	2.65 ± 0.04	0.96 ± 0.04
	3.47 mM Cr(VI)	0	1.05 ± 0.04	2.40 ± 0.02	0	1.19 ± 0.01
	3.47 mM Cr(VI) (wb)	0	3.34 ± 0.05	0.12 ± 0.01	0	0

Notes: (wb) – the media without bacteria; to media with NO₂⁻ and Cr(VI) or without it the NH₄Cl was not added.

In the media with NO_2^- and 10.41 mM Cr(VI) the growth of bacteria decreased 2.2–2.3 times, compared with growth in the media with NaNO_2 as the sole electron acceptor. In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increase of Cr(VI) concentrations there was a gradual (1.4–2.8 times) decrease in the efficiency of nitrite ions' reduction by bacteria, as compared with their reduction in the media with NaNO_2 (92.8–94.8%, Fig. 3a). In the media, containing NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$, the cells produced 1.15–2.25 mM of ammonium ions (control – 2.53–2.59 mM, Table 3). The efficiency of Cr(VI) reduction by bacteria with increase in its concentration in the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ was

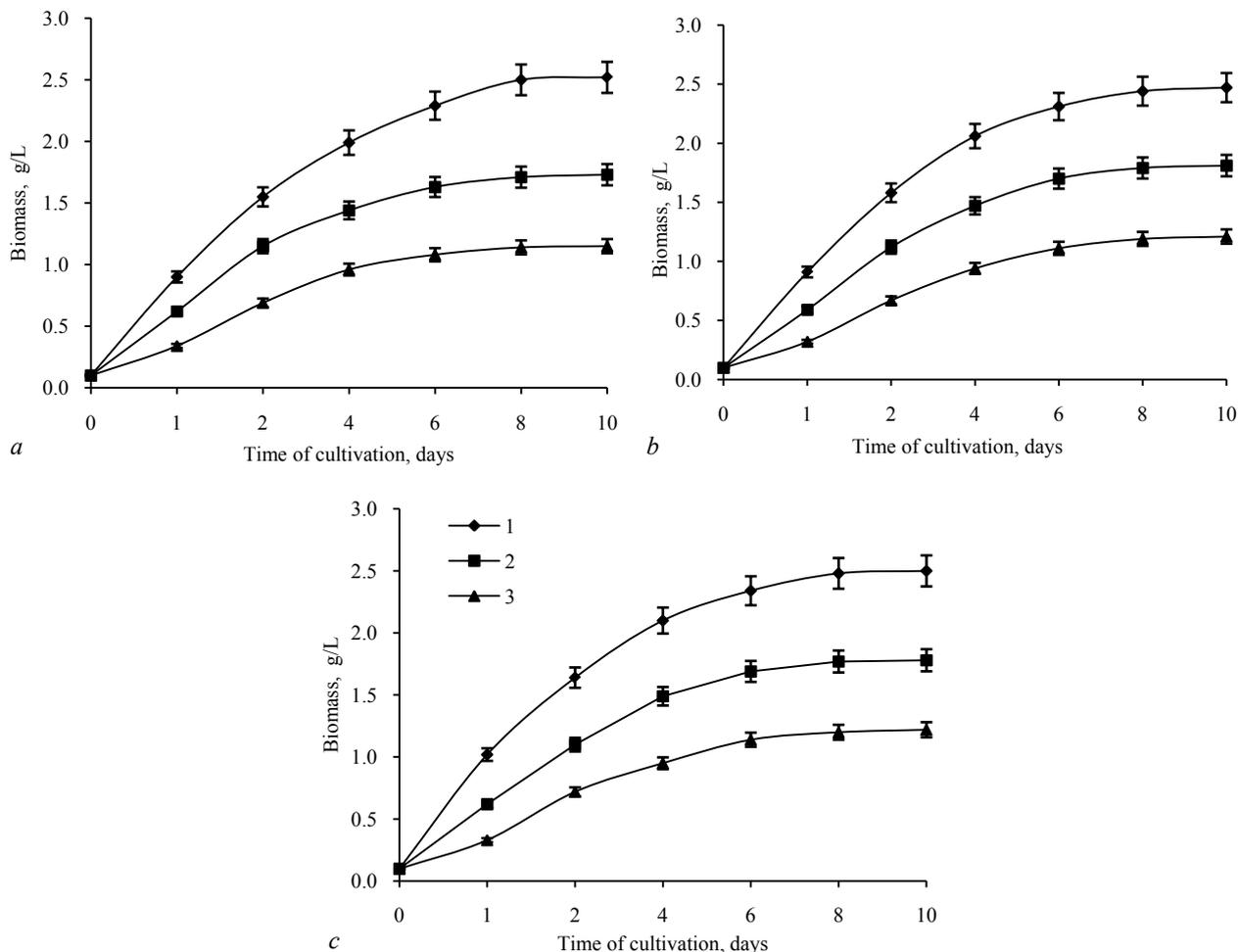


Fig. 6. Biomass accumulation by *Desulfuromonas acetoxidans* IMV B-7384 (a), *Desulfuromonas sp.* Yavor-5 (b) and *Desulfuromonas sp.* Yavor-7 (c) during growth in media with NaNO_3 (1), NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ (2), $\text{K}_2\text{Cr}_2\text{O}_7$ (3) ($x \pm \text{SD}$, $n = 3$)

In the media with the same initial content of (3.47 mM) NO_2^- and Cr(VI) the reduction of Cr(VI) by bacteria was only slightly lower (up to 1.1 times) than the reduction of nitrite ions, the almost same concentrations of trivalent chromium and ammonium ions were detected in the cultural liquid.

Sulfur reducing bacteria play an important role in regulating the level not only of sulfur and carbon compounds, but also of nitrogen and metals in the environment. Contamination of the environment with heavy metals adversely affects the physiological and biochemical processes carried out by these bacteria. The features of the metabolism of microorganisms under the influence of toxic nitrogen compounds (in particular, nitrates and nitrites) as stressors often remains poorly understood. Therefore, we studied the ability of *Desulfuromonas sp.* bacteria to reduce in the process of anaerobic respiration nitrate or nitrite ions at different concentrations with the simultaneous presence of Cr(VI) in the media.

The influence of 3.47 mM Cr(VI) on the 1.74–10.41 mM nitrate ions usage by bacteria was studied. Bacteria were cultivated in the media without NH_4Cl , to which NaNO_3 at different concentrations and $\text{K}_2\text{Cr}_2\text{O}_7$ at Cr(VI) concentration of 3.47 mM were added. The bacteria were also sown in the media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final NO_3^- or Cr(VI) con-

centration in the media of 3.47 mM, to test the bacteria growth in the media with nitrate ions or hexavalent chromium as the sole electron acceptor (Table 4). After 10 days of growth the biomass of bacteria in the media with NaNO_3 was found to be 2.0–2.2 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$. After simultaneous addition into the media of NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increasing concentrations of NO_3^- a gradual inhibition of bacteria growth was observed, compared with growth in the media with only NaNO_3 . In the media with 10.41 mM NO_3^- and Cr(VI) the growth of bacteria decreased up to 2.5 times, compared with the growth in the media with NaNO_3 as the sole electron acceptor. In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increase in NO_3^- concentrations there was also a gradual (1.2–3.6 times) decrease in the efficiency of nitrate ions' reduction by cells, compared with their reduction in the media with only NaNO_3 (90.5–92.2%, Fig. 4a). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 1.26–2.95 mM of ammonium ions (control – 2.69–2.73 mM, Table 4). The efficiency of Cr(VI) reduction by cells with increasing NO_3^- concentrations in the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ was revealed to be 1.1–1.6 times lower than its reduction in the media only with $\text{K}_2\text{Cr}_2\text{O}_7$ (68.6–70.3%, Fig. 4b). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 1.49–2.12 mM of Cr(III) (control – 2.36–2.43 mM, Table 4).

revealed to be from 1.2 to 2.7 times lower than its reduction in the media with $\text{K}_2\text{Cr}_2\text{O}_7$ (69.7–72.0%, Fig. 3b). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ cells produced 0.89–3.09 mM of the Cr(III) (control – 2.37–2.42 mM, Table 3).

In the media with NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ without bacteria the reduction of NO_2^- and Cr(VI) did not exceed 3.8% (Fig. 3). Thus, it has been established that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits the biomass accumulation, the nitrite ions' reduction and the ammonium ions' production by bacteria of *Desulfuromonas sp.* after simultaneous addition into the media of 3.47 mM NO_2^- and Cr(VI) (1.74–10.41 mM).

In media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ without bacteria the efficiency of NO_3^- and Cr(VI) reduction did not exceed 3.5% and 4.0%, respectively (Fig. 4). Thus, it has been shown that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits the biomass accumulation, the nitrate ions' reduction and the ammonium ions' production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 1.74–10.41 mM NO_3^- and 3.47 mM Cr(VI) . In the media with the same initial content (3.47 mM) of NO_3^- and Cr(VI) bacteria reduced up to 1.2 times more nitrate ions than Cr(VI) with the production of ammonium ions at concentrations the same times higher than that of Cr(III) .

The usage of 1.74–10.41 mM nitrite ions by bacteria under the influence of 3.47 mM Cr(VI) was studied. Bacteria were cultivated in media without NH_4Cl , to which NaNO_2 at different concentrations and 3.47 mM Cr(VI) in form of $\text{K}_2\text{Cr}_2\text{O}_7$ were added. The bacteria were also sown in media with NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final NO_2^- or Cr(VI) concentration in the media of 3.47 mM, to test the bacteria growth in the media with nitrite ions or hexavalent chromium as the sole electron acceptor (Table 5). Biomass of bacteria in the media with NaNO_2 was revealed to be 1.9–2.0 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$. After simultaneous addition into the media of NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increasing concentrations of NO_2^- there was a decrease in the bacteria growth, compared with growth in a media with NaNO_2 . In the media with 10.41 mM NO_2^- and Cr(VI) the growth of bacteria decreased 2.4–2.6 times, compared with growth in

media with only NaNO_2 . In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ with increase in NO_2^- concentrations there was a gradual (1.4–3.9 times) decrease in the efficiency of nitrite ions' reduction by bacteria, as compared with their reduction in the media with NaNO_2 (92.5–94.5%, Fig. 5a). In the media containing NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$, the cells produced 1.10–2.65 mM of ammonium ions (control – 2.53–2.59 mM, Table 5). The efficiency of the Cr(VI) reduction by bacteria with increase in NO_2^- concentrations in the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ was revealed to be from 1.2 to 1.7 times lower than its reduction in the media with $\text{K}_2\text{Cr}_2\text{O}_7$ (69.7–72.0%, Fig. 5b). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ cells produced 1.48–2.07 mM of the Cr(III) (control – 2.40–2.48 mM, Table 5).

In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ without bacteria the reduction of NO_2^- and Cr(VI) did not exceed 4.0 and 3.8%, respectively (Fig. 5). Thus, it has been established that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits the biomass accumulation, the nitrite ions' reduction and the ammonium ions' production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 1.74–10.41 mM NO_2^- and 3.47 mM Cr(VI) . In the media with the same initial content (3.47 mM) NO_2^- and Cr(VI) the reduction of Cr(VI) by bacteria was only slightly, up to 1.1 times, lower than the reduction of nitrite ions, almost the same concentrations of trivalent chromium and ammonium ions were detected in the cultural liquid.

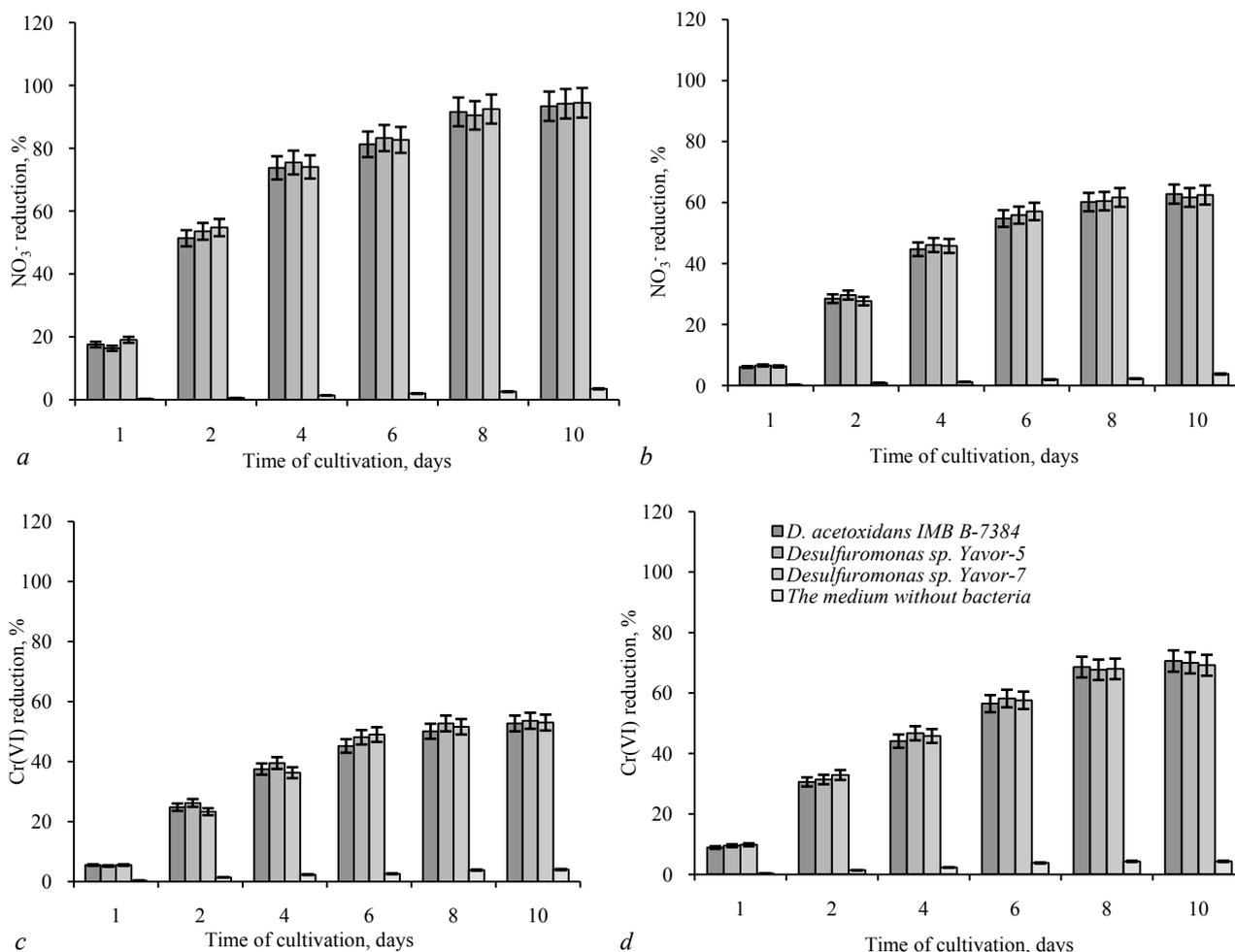


Fig. 7. Efficiency of 3.47 mM NO_3^- (a, b) or 3.47 mM Cr(VI) (c, d) reduction by *Desulfuromonas* sp. after 10 days of growth in the media with NaNO_3 (a), NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ (b, c), $\text{K}_2\text{Cr}_2\text{O}_7$ (d) and in the same media without bacteria ($x \pm \text{SD}$, $n = 3$)

Bacteria were grown during 10 days in the media without NH_4Cl , to which NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ were added to equal NO_3^- and Cr(VI) concentrations in the media of 3.47 mM. The bacteria were also sown in the media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final NO_3^- or Cr(VI) concentration in the media of 3.47 mM, to test the bacteria growth in the media with nitrate ions or hexavalent chromium as the sole electron acceptor (Fig. 6–8). On day 10 of growth the biomass of bacteria in the media

with NaNO_3 was revealed to be 2.0–2.2 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$. In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ the biomass of bacteria was 1.4–1.5 times lower than in the media with only NaNO_3 , but 1.5 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$ as the sole electron acceptor (Fig. 6). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ there was a 1.5 times decrease in the efficiency of nitrate ions reduction by cells, compared with their reduction in the media with only NaNO_3 (93.4–

94.5%, Fig. 7a, b). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 2.11–2.13 mM of ammonium ions (control – 2.45–2.49 mM) (Fig. 8a, b, c). The efficiency of Cr(VI) reduction by cells in the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ was revealed to be up to 1.3 times lower than its reduction in the media only with $\text{K}_2\text{Cr}_2\text{O}_7$ (69.2–70.6%, Fig. 7c, d). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 1.79–1.83 mM of Cr(III) (control – 2.36–2.40 mM, Fig. 8d, e, f). In the media with NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria reduced 1.2 times more nitrate ions

than Cr(VI) with the production of NH_4^+ at concentrations 1.2 times higher than that of Cr(III) (Fig. 8). In media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ without bacteria the efficiency of NO_3^- and Cr(VI) reduction did not exceed 3.5–3.8% and 4.0–4.3%, respectively (Fig. 7). Thus, it has been shown that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits the biomass accumulation, the nitrate ions' reduction and the ammonium ions' production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 3.47 mM NO_3^- and 3.47 mM Cr(VI).

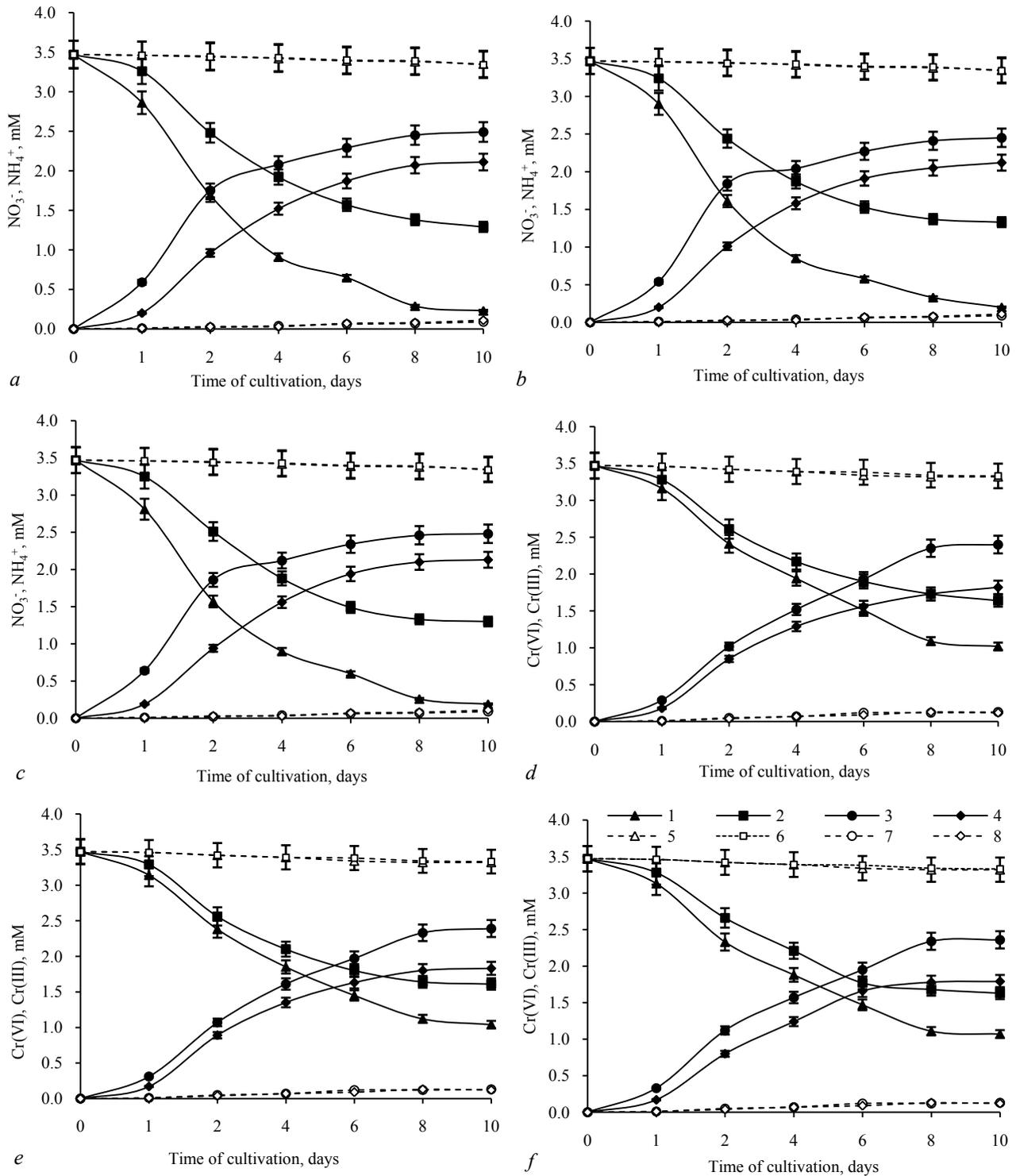


Fig. 8. Residual content of NO_3^- or Cr(VI) (1, 2, 5, 6) and concentrations of NH_4^+ or Cr(III) (3, 4, 7, 8) in cultural liquid during growth of *Desulfuromonas acetoxidans* IMV B-7384 (a, d), *Desulfuromonas* sp. Yavor-5 (b, e) and *Desulfuromonas* sp. Yavor-7 (c, f) in the media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ (1, 3), NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ (2, 4) and in the media with NaNO_3 or $\text{K}_2\text{Cr}_2\text{O}_7$ (5, 7), NaNO_3 and $\text{K}_2\text{Cr}_2\text{O}_7$ (6, 8) without bacteria ($x \pm \text{SD}$, $n = 3$)

Bacteria were cultivated during 10 days in the media without NH_4Cl , to which NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ were added to equal NO_2^- and Cr(VI) concentrations in the media of 3.47 mM. The bacteria were also sown in the media with NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ to final NO_2^- or Cr(VI) concentration in the media of 3.47 mM, to test the bacteria growth in the media with nitrite ions or hexavalent chromium as the sole electron acceptor (Fig. 9–11). On day 10 of growth the biomass of bacteria in the media with NaNO_2 was revealed to be 1.9–2.1 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$. In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ the biomass of bacteria was 1.4–1.5 times lower than in the media with only NaNO_2 , but 1.3–1.4 times higher than in the media with $\text{K}_2\text{Cr}_2\text{O}_7$ as the sole electron acceptor (Fig. 9). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ there was a 1.7–1.8 times decrease in the efficiency of nitrite ions' reduction by cells, compared with their reduction in the media with only NaNO_2 (93.7–94.8%, Fig. 10a, b). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 1.76–1.87 mM

of ammonium ions (control – 2.30–2.37 mM, Fig. 11a, b, c). The efficiency of Cr(VI) reduction by cells in the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ was revealed to be 1.4–1.5 times lower than its reduction in the media only with $\text{K}_2\text{Cr}_2\text{O}_7$ (69.9–72.2%, Fig. 10c, d). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria produced 1.63–1.71 mM of Cr(III) (control – 2.42–2.49 mM, Fig. 11d, e, f). In the media with NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ bacteria reduced up to 1.1 times more nitrite ions than Cr(VI) with the production of NH_4^+ at concentrations the same times higher than that of Cr(III) (Fig. 11). In media with NaNO_2 or $\text{K}_2\text{Cr}_2\text{O}_7$ without bacteria the efficiency of NO_2^- and Cr(VI) reduction did not exceed 3.5–3.8% and 4.3–4.6%, respectively (Fig. 10). Thus, it has been shown that $\text{K}_2\text{Cr}_2\text{O}_7$ inhibits the biomass accumulation, the nitrite ions' reduction and the ammonium ions' production by bacteria of *Desulfuromonas* sp. after simultaneous addition into the media of 3.47 mM NO_2^- and 3.47 mM Cr(VI) .

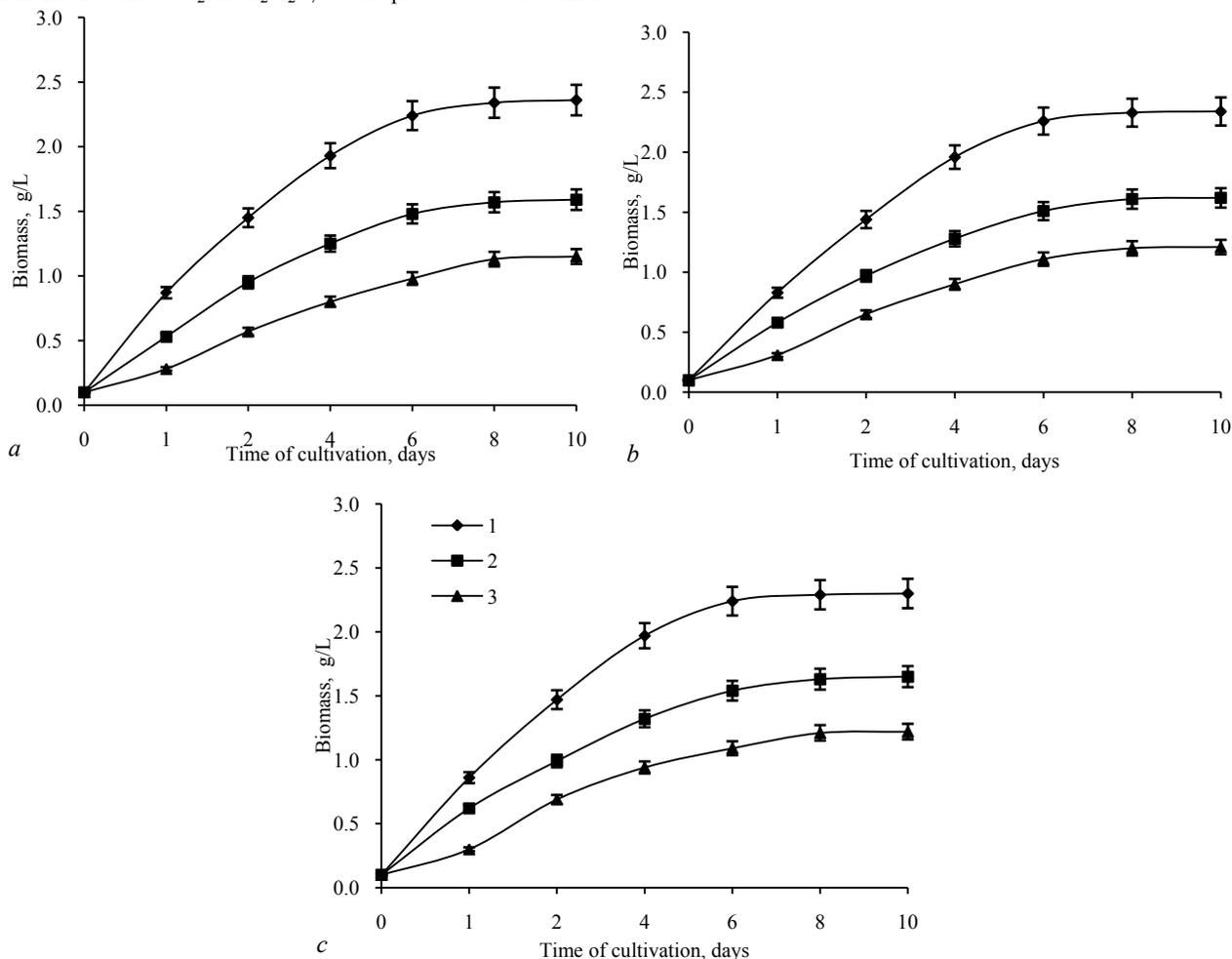


Fig. 9. Biomass accumulation by *Desulfuromonas acetoxidans* IMV B-7384 (a), *Desulfuromonas* sp. Yavor-5 (b) and *Desulfuromonas* sp. Yavor-7 (c) during growth in media with NaNO_2 (1), NaNO_2 and $\text{K}_2\text{Cr}_2\text{O}_7$ (2), $\text{K}_2\text{Cr}_2\text{O}_7$ (3) ($x \pm \text{SD}$, $n = 3$)

Discussion

As a result of the constant ingress into the environment of various chemical contaminants at critical concentrations, the metabolism of bacteria slows down, the species composition of microbiocenoses changes, and in the process of natural selection few resistant strains of microorganisms survive. Sulfur reducing bacteria of the *Desulfuromonas* genus, isolated by us from the man-made Yavorivske Lake, in the process of oxidation of organic compounds use, besides sulfur, oxidized forms of nitrogen and heavy metals, in particular, Cr(VI) as electron acceptors of anaerobic respiration. The intensity of anaerobic respiration in these microorganisms in contaminated ecotopes is determined by level of their adaptation to unfavourable environmental conditions, in particular, increased content of metal compounds (Viti et al., 2014; Simonte et al., 2017; Teng et al., 2019). Therefore, the ability of bacteria of *Desulfuromonas* sp. to reduce

sulfur, nitrate or nitrite ions at a simultaneous presence in the media of $\text{K}_2\text{Cr}_2\text{O}_7$ at Cr(VI) concentrations 1.74–10.41 mM (0.5–3.0 times different from the standard electron acceptor content in conventional for sulfur reducing bacteria Kravtsov-Sorokin media) was studied.

The efficiency of electron acceptor reduction by bacteria is primarily determined by the difference between the oxidation-reduction potential of the electron donor and acceptor, which depends on the pH of the media and changes during cultivation of bacteria (Gescher & Kappler, 2012; Govorukha et al., 2015). Bacteria of the *Desulfuromonas* genus oxidize simple organic substrates completely to CO_2 and H_2O in the tricarboxylic acid cycle or in the acetyl-CoA/CO-dehydrogenase pathway (Sung et al., 2003; An & Picarda, 2015; Vasylyv et al., 2015). Although the succession of electron acceptors' reduction by microorganisms at their simultaneous presence in the media is determined by their standard oxidation-reduction potential (pH 7.0), the energy supply of cells during anaerobic respiration

depends on the ways of ATP synthesis in the process of electron donor oxidation: by substrate or oxidative phosphorylation (Lengeler et al., 2005; McKinlay et al., 2020). In natural environments there usually are several possible electron acceptors of anaerobic respiration, and bacteria first of all reduce the most profitable acceptors for them. In every microorganism the succession of use of different electron acceptors is determined genetically and controlled by complex regulatory mechanisms (Lengeler et al., 2005; Kozlova et al., 2008; Rosenberg et al., 2014).

At simultaneous presence in the cultivation media of sulfur and $K_2Cr_2O_7$ bacteria reduce more Cr(VI) than S^0 (at pH 7.0 the standard oxidation-reduction potential ($E_0' = +1.33$ V) of the Cr(VI)/Cr(III) pair is considerably higher than that ($E_0' = -0.27$ V) of S^0/HS^- pair) (Lengeler et al., 2005; An & Picarda, 2015). The evidence of this is the fact that in the media with the same (3.47 mM) initial concentration of S^0 and Cr(VI) the content of H_2S was 3.3–3.4 times lower than the content of Cr(III), produced by bacteria. In the sulfur respiration of bacteria of *Desulfuromonas* genus sulfur reductase and polysulfide reductase are involved, which are located in the cytoplasmic membrane and bounded with hydrogenase

by components of the electron transport chain (Lengeler et al., 2005; Chayka et al., 2016). Cr(VI) at all tested concentrations in the media represses the dissimilatory sulfur reduction carried out by bacteria, as evidenced the decrease in the concentrations of produced by bacteria hydrogen sulfide in the media with S^0 and $K_2Cr_2O_7$.

Under these conditions of growth Cr(VI) may damage the structure of the cytoplasmic membrane of bacteria and thus influence on the activity of membrane-bound enzymes (Maslovska & Hnatush, 2013). Although the reduction of metals-oxidants by membrane-bound metal reductases is mainly carried out outside the cell (Gescher & Kappler, 2012; Richter et al., 2012; Simonte et al., 2017), with increase in the concentration of soluble $K_2Cr_2O_7$ in the media, increase in the degree of Cr(VI) penetration through the cytoplasmic membrane of bacteria into the cytoplasm, where its interaction with intracellular metabolites occurs, oxygen radicals are formed, Cr(III) accumulates as a reduced end product, which causes the inhibition of growth and metabolic activity of bacteria (Richter et al., 2012; Viti et al., 2014; Hnatush & Maslovska, 2018).

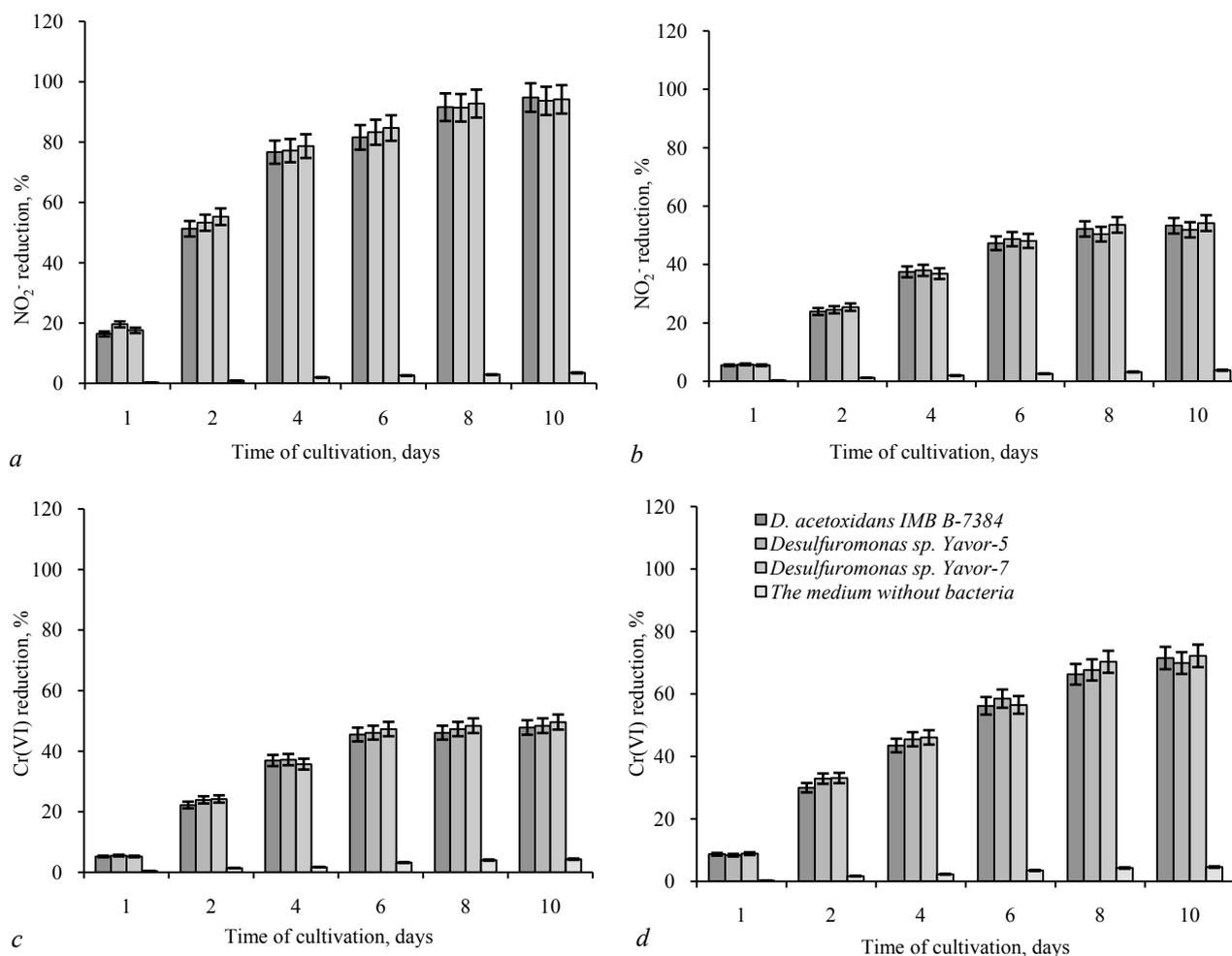


Fig. 10. Efficiency of 3.47 mM NO_2^- (a, b) or 3.47 mM Cr(VI) (c, d) reduction by *Desulfuromonas* sp. after 10 days of growth in the media with $NaNO_2$ (a), $NaNO_2$ and $K_2Cr_2O_7$ (b, c), $K_2Cr_2O_7$ (d) and in the same media without bacteria ($x \pm SD$, $n = 3$)

Despite the fact that the standard redox potential at pH 7.0 of the Cr(VI)/Cr(III) pair ($E_0' = +1.33$ V) is considerably higher than that of NO_3^-/NO_2^- and NO_2^-/NH_4^+ pairs ($E_0' = +0.43$ and $+0.34$ V, respectively) (Lengeler et al., 2005; Kozlova et al., 2008; Richter et al., 2012), in the media with the same initial content (3.47 mM) of NO_3^- or NO_2^- and Cr(VI) bacteria reduced more nitrate and nitrite ions than Cr(VI). Nevertheless, $K_2Cr_2O_7$ inhibited the nitrate and nitrite reduction, which bacteria carried out, at all investigated concentrations of inorganic toxicants in the media. The negative influence of Cr(VI) on the activity of molybdenum-containing membrane-bound dissimilatory nitrate reductase (Morozkina & Zvyagilskaya, 2007), as well as periplasmic nitrite reductase, containing

siroheme as a prosthetic group (Lengeler et al., 2005), in bacteria of *Desulfuromonas* genus can be caused by damage to the structure of cytoplasmic membrane or modification of the active conformation and denaturation of molecules of these enzymes. Sulfur, nitrates and nitrites are less-powerful electron acceptors of anaerobic respiration to sulfidogenic bacteria than oxidized forms of metals (Cadby et al., 2017). But they use a thermodynamically more favourable electron acceptor, such as hexavalent chromium, to a lesser extent, possibly due to its high toxicity.

Although the reduction of 1.74–10.41 mM Cr(VI) by cells in media with 3.47 mM S^0 , $NaNO_3$ or $NaNO_2$ and $K_2Cr_2O_7$ decreased by 1.1–2.0, 1.2–2.3 and 1.2–2.7 times, respectively, compared with its reduction in

media only with $K_2Cr_2O_7$, anaerobic respiration by bacteria was performed quite effectively using different electron acceptors.

In the media with the same initial content (3.47 mM) of NO_3^- or NO_2^- and Cr(VI) bacteria produced 1.1–1.2 times more NH_4^+ than Cr(III), but in the media with the same initial content (3.47 mM) of S^0 and Cr(VI) bacteria produced more than three times Cr(III) than hydrogen sulfide. The obtained data indicate that the processes of nitrate and nitrite reduction, carried out by bacteria, are less sensitive to the negative influence

of sodium dichromate, than reduction of sulfur. Due to the ability to use different electron acceptors and reduce them at high concentrations (up to 10.41 mM) adapted to survival conditions the investigated strains of bacteria can be the basis of the new developments in the field of biotechnology.

Our data allow us to conclude that bacteria of *Desulfuromonas* genus play an important role in the geochemical cycles of sulfur, nitrogen and chromium in technogenically altered ecotopes.

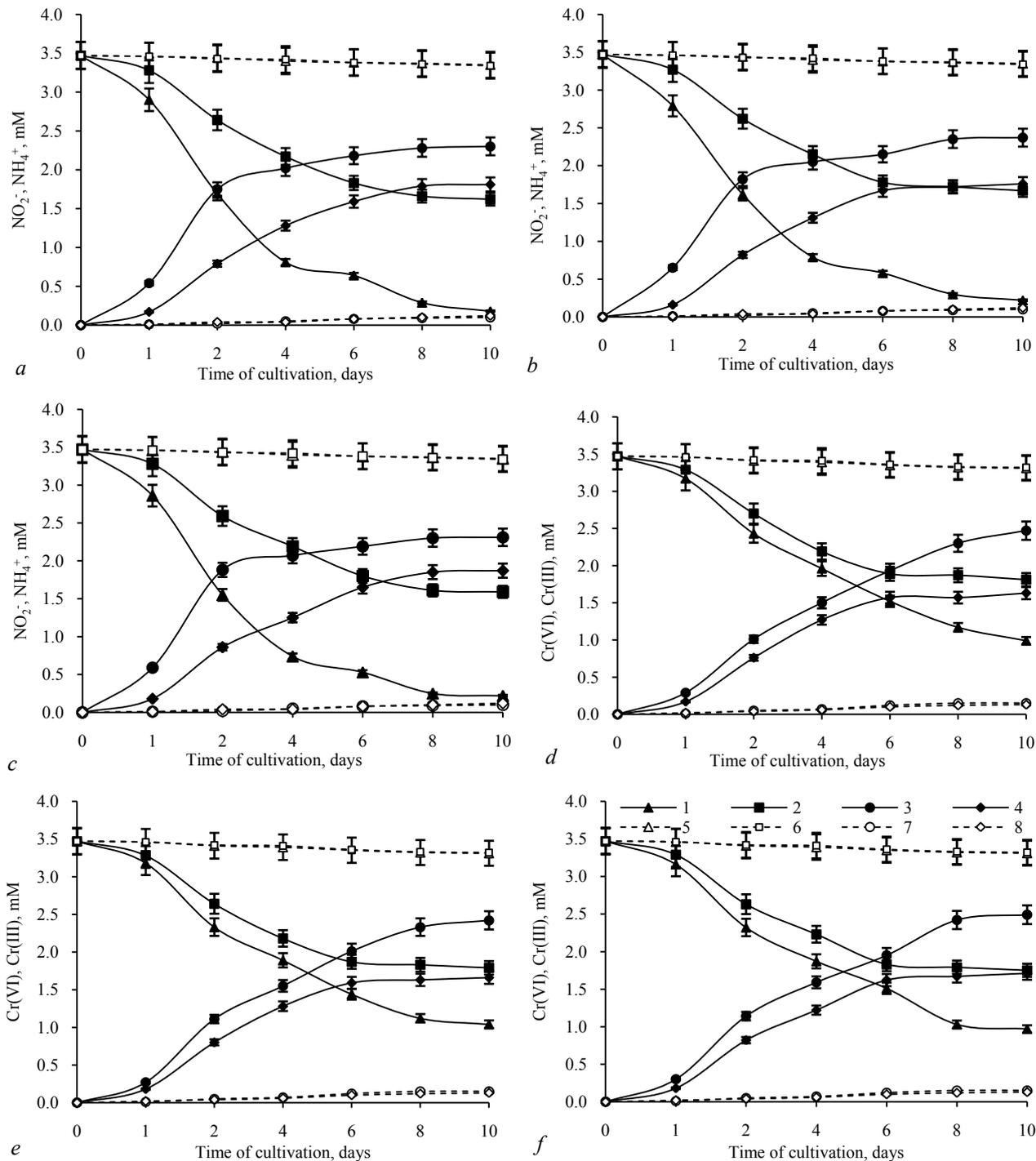


Fig. 11. Residual content of NO_2^- or Cr(VI) (1, 2, 5, 6) and concentrations of NH_4^+ or Cr(III) (3, 4, 7, 8) in cultural liquid during growth *Desulfuromonas acetoxidans* IMV B-7384 (a, d), *Desulfuromonas* sp. Yavor-5 (b, e) and *Desulfuromonas* sp. Yavor-7 (c, f) in the media with $NaNO_2$ or $K_2Cr_2O_7$ (1, 3), $NaNO_2$ and $K_2Cr_2O_7$ (2, 4) and in the media with $NaNO_2$ or $K_2Cr_2O_7$ (5, 7), $NaNO_2$ and $K_2Cr_2O_7$ (6, 8) without bacteria ($x \pm SD$, $n = 3$)

Conclusion

Thus, it was established that bacteria of *Desulfuromonas* genus, isolated from Yavorivske Lake, use sulfur, nitrates, nitrites and Cr(VI) as electron acceptors in the process of anaerobic respiration at concentrations

of 3.47 mM or 1.74–10.41 mM in the media. Cr(VI) at all tested concentrations inhibits the hydrogen sulfide production by bacteria in the media with S^0 and $K_2Cr_2O_7$. Cr(VI) at all concentrations negatively affects the nitrate or nitrite ions' reduction and the ammonium ions' production by bacteria in the media with $NaNO_3$ or $NaNO_2$ and $K_2Cr_2O_7$. Nevertheless,

the efficiency of Cr(VI) reduction by cells in the media with 3.47 mM S⁰, NaNO₃ or NaNO₂ and K₂Cr₂O₇ at Cr(VI) concentration of 10.41 mM was not lower than 35.9%, 30.1% and 26.6%, respectively. The studied strains are capable of reductive transformation of inorganic toxicants at high concentrations (up to 10.41 mM) and therefore are promising for use in technologies for purifying environments with complex pollution from chromium compounds.

The work was financially supported by the Ministry of Education and Science of Ukraine "Modelling and prediction of the influence of chemical contaminants on microorganisms, which convert sulfur compounds", No. 0121U109616.

References

- Aguilar-Barajas, E., Diaz-Perez, C., Ramirez-Diaz, M. I., Riveros-Rosas, H., & Cervantes, C. (2011). Bacterial transport of sulfate, molybdate, and related oxyanions. *Biometals*, 24, 687–707.
- An, T. T., & Picarda, F. W. (2015). *Desulfuromonas carbonis* sp. nov., an Fe (III)-, S⁰ and Mn (IV)-reducing bacterium isolated from an active coalbed methane gas well. *International Journal of Systematic and Evolutionary Microbiology*, 65(5), 1686–1693.
- Belchik, S. M., Kennedy, D. W., Dohnalkova, A. C., Wang, Y. M., Sevinc, P. C., Wu, H., Lin, Y. H., Lu, H. P., Fredrickson, J. K., & Shi, L. (2011). Extracellular reduction of hexavalent chromium by cytochromes MtrC and OmcA of *Shewanella oneidensis* MR-1. *Applied and Environmental Microbiology*, 77(12), 4035–4041.
- Bilyy, O. I., Vasylyv, O. M., & Hnatysh, S. O. (2014). The anode biocatalyst with simultaneous transition metals pollution control. Technology and application of microbial fuel cells. InTech, Rijeka.
- Bokranz, M. J., Katz, J., Schröder, I., Robertson, A. M., & Kröger, A. (1983). Energy metabolism and biosynthesis of *Vibrio succinogenes* growing with nitrate or nitrite as terminal electron acceptor. *Archives of Microbiology*, 135, 36–41.
- Breuer, M., Rosso, K. M., Blumberger, J., & Butt, J. N. (2015). Multi-haem cytochromes in *Shewanella oneidensis* MR-1: Structures, functions and opportunities. *Journal of the Royal Society Interface*, 12(102), 20141117.
- Caballero-Flores, G. G., Costa-Navarrete, Y. M., Ramirez-Diaz, M. I., Silva-Sanchez, J., & Cervantes, C. (2012). Chromate-resistance genes in plasmids from antibiotic-resistant nosocomial enterobacterial isolates. *FEMS Microbiology Letters*, 327(2), 148–154.
- Cadby, I. T., Faulkner, M., Cheneby, J., Long, J., van Helden, J., Dolla, A., & Cole, J. A. (2017). Coordinated response of the *Desulfovibrio desulfuricans* 27774 transcriptome to nitrate, nitrite and nitric oxide. *Scientific Reports*, 7, 16228.
- Chayka, O. M., & Peretyatko, T. B. (2018). The reduction of hexavalent chromium and nitrates by *Desulfuromonas* sp. YSDS-3, isolated from the soil of Yasiv sulfur mine. *Ecology and Noospherology*, 29(2), 76–82.
- Chayka, O., Peretyatko, T., Gudz, S., & Hnatysh, S. (2016). Sulfur reducing activity of the *Desulfuromonas acetoxidans* IMV B-7384 under different cultivation conditions. *Visnyk of Lviv University, Biological Series*, 74, 161–168.
- Fathima, N. N., Aravindhan, R., Rao, J. R., & Nair, B. U. (2005). Solid waste removes toxic liquid waste: Adsorption of chromium (VI) by iron complexed protein waste. *Environmental Science and Technology*, 39(8), 2804–2810.
- Fitzgerald, L. A., Petersen, E. R., Leary, D. H., Nadeau, L. J., Soto, C. M., Ray, R. L., Little, B. J., Ringeisen, B. R., Johnson, G. R., Vora, G. J., & Biffinger, J. C. (2013). *Shewanella frigidimarina* microbial fuel cells and the influence of divalent cations on current output. *Biosensors and Bioelectronics*, 40(1), 102–109.
- Gescher, J., & Kappler, A. (2012). Microbial metal respiration: From geochemistry to potential applications. Springer-Verlag, Berlin, Heidelberg.
- Govorukha, V. M., Havrylyuk, O. A., & Tashyrev, O. B. (2015). Regularities of quantitative distribution for Fe(III)-reducing bacteria in natural ecosystems. *Biotechnologia Acta*, 8(3), 123–128.
- Granger, D. L., Taintor, R. R., Boockvar, K. S., & Hibbs, J. B. (1996). Measurement of nitrate and nitrite in biological samples using nitrate reductase and Griess reaction. *Methods Enzymology*, 268, 142–151.
- Gudz, S. P., Hnatysh, S. O., Yavorska, G. V., Bilinska, I. S., & Borsukevych, B. M. (2014). Praktikum z mikrobiologii [Workshop on microbiology]. Ivan Franko National University of Lviv, Lviv (in Ukrainian).
- Hedderich, R., Klimmek, O., Kroger, A., Dirmeier, R., Keller, M., & Stetter, K. O. (1999). Anaerobic respiration with elemental sulfur and with disulfides. *FEMS Microbiology Reviews*, 22(5), 353–381.
- Hnatysh, S. O., Moroz, O. M., Yavorska, G. V., & Borsukevych, B. M. (2018). Sulfidogenic and metal reducing activities of *Desulfuromonas* genus bacteria under the influence of copper chloride. *Biosystems Diversity*, 26(3), 218–226.
- Hnatysh, S., & Maslovska, O. (2018). Sulfur-reducing bacteria *Desulfuromonas acetoxidans* IMV B-7384 under the influence of heavy metal ions. The Development of Natural Sciences. Baltija Publishing, Riga.
- Hoffmann, M. C., Pfänder, Y., Tintel, M., & Masepohl, B. (2017). Bacterial PerO permeases transport sulfate and related oxyanions. *Journal of Bacteriology*, 199(14), e00183-17.
- Jing, X., Wu, Y., Shi, L., Peacock, C. L., Ashry, N. M., Gao, C., Huang, Q., & Cai, P. (2020). Outer membrane c-type cytochromes OmcA and MtrC play distinct roles in enhancing the attachment of *Shewanella oneidensis* MR-1 cells to goethite. *Applied and Environmental Microbiology*, 86(23), e01941-20.
- Joutey, N. T., Sayel, H., Bahafid, W., & El Ghachtouli, N. (2015). Mechanisms of hexavalent chromium resistance and removal by microorganisms. *Reviews of Environmental Contamination and Toxicology*, 233, 45–69.
- Kazakis, N., Kougiyas, I., & Patsialis, T. (2015). Assessment of flood hazard areas at a regional scale using an index-based approach and analytical hierarchy process: Application in Rhodope-Evros Region, Greece. *Science of the Total Environment*, 538, 555–563.
- Kiran, M. G., Pakshirajan, K., & Das, G. (2017). Heavy metal removal from multi-component system by sulfate reducing bacteria: Mechanism and cell surface characterization. *Journal of Hazardous Materials*, 324(A), 62–70.
- Kozlova, I. P., Radchenko, O. S., Stepura, L. H., Kondratyuk, T. O., & Pilyashenko-Novokhatnyy, A. I. (2008). Heokhimichna diyalnist mikroorganizmiv ta yivy prykladni aspekty [Geochemical activity of microorganisms and its applied aspects]. *Naukova Dumka, Kyiv* (in Ukrainian).
- Kuever, J., Rainey, F. A., & Widdel, F. (2005). Family I. Desulfuromonaceae fam. nov. Genus I. *Desulfuromonas* / *Desulfuromonas* genus. Pfennig and Biebl, 1977. In: Brenner, D. J., Krieg, N. R., Staley, J. T., & Garrity, G. M. (Eds.). *Bergey's manual of systematic bacteriology*. Vol. 2. Springer, New York.
- Kuznetsov, A., Gradova, N., Lushnikov, S., Engelkhart, M., Vaysser, T., & Chebotareva, M. (2015). *Prikladnaya ekhbiotekhnologiya* [Applied Ecobiotechnology]. Binom, Moscow (in Russian).
- Lengeler, J., Dreves, G., & Shlegel, G. (Eds.). (2005). *Sovremennaya mikrobiologiya. Prokarioty* [Contemporary Microbiology. Prokaryotes]. Mir, Moscow (in Russian).
- Liang, J., Huang, X., Yan, J., Li, Y., Zhao, Z., Liu, Y., Ye, J., & Wei, Y. (2021). A review of the formation of Cr(VI) via Cr(III) oxidation in soils and groundwater. *Science of the Total Environment*, 774, 145762.
- Mandich, N. V. (1997). Chemistry and theory of chromium deposition: Part I – Chemistry. *Plating and Surface Finishing*, 84(5), 108–115.
- Maslovska, O. D., & Hnatysh, S. O. (2013). Vplyv ferum (III) cytratu na ATF-gidrolazy *Desulfuromonas acetoxidans* IMV B-7384 [The influence of ferric (III) citrate on ATP-hydrolases of *Desulfuromonas acetoxidans* IMV B-7384]. *Biosystems Diversity*, 21(1), 3–8 (in Ukrainian).
- Maslovska, O., Hnatysh, S., & Katemyak, S. (2015). The activity of enzymes of glutathione antioxidant system of *Desulfuromonas acetoxidans* IMV B-7384 under the influence of ferric (III) citrate. *Visnyk of Lviv University, Biological Series*, 70, 213–220.
- McKinlay, J. B., Cook, G. M., & Hards, K. (2020). Microbial energy management – a product of three broad tradeoffs. *Advances in Microbial Physiology*, 77, 139–185.
- Moroz, O. M., Hnatysh, S. O., Bohoslavets, C. I., Yavorska, G. V., & Truchym, N. V. (2016). Vykorystannya bakteriyamy *Desulfuromonas* sp. yoniv ferumu (III) i manhanu (IV) yak akseptoriv elektroniv [Usage of ferum (III) and manganese (IV) ions as electron acceptors by bacteria of *Desulfuromonas* sp.]. *Biosystems Diversity*, 24(1), 87–95 (in Ukrainian).
- Moroz, O. M., Hnatysh, S. O., Tarabas, O. V., Bohoslavets, C. I., Yavorska, G. V., & Borsukevych, B. M. (2018). Sulfidogenna aktyvnist sulfatvidnovlyvalnyh i sirkovidnovlyvalnyh bakteriy za vplyvu spoluk metaliv [Sulfidogenic activity of sulfate and sulfur reducing bacteria under the influence of metal compounds]. *Biosystems Diversity*, 26(1), 3–10 (in Ukrainian).
- Moroz, O. M., Peretyatko, T. B., Klym, I. R., Borsukevych, B. M., Yavorska, G. V., & Kulachkovsky, O. R. (2013). Sirkovidnovlyvalni bakteriyi ozera Yavorivske: Deyaki morfolohichni, kulturalni i fiziolohichni osoblyvosti [Sulfur reducing bacteria from Yavorivske lake: Some morphological, cultural and physiological peculiarities]. *Scientific Bulletin of the Uzhgorod University, Series Biology*, 35, 34–41 (in Ukrainian).
- Moroz, O., Gul, N., Galushka, A., Zvir, G., & Borsukevych, B. (2014). Vykorystannya riznyh akceptoriv elektroniv bakteriyamy *Desulfuromonas* sp., vydilennyh z ozera Yavorivske [Different electron acceptors usage by bacteria of *Desulfuromonas* sp. isolated from Yavorivske Lake]. *Visnyk of Lviv University, Biological Series*, 65, 322–334 (in Ukrainian).
- Morozkina, E. V., & Zvyagilskaya, R. A. (2007). Nitrate reductases: Structure, functions, and effect of stress factors. *Biochemistry*, 72(10), 1151–1160.
- Mustapha, M. U., & Halimoon, N. (2015). Screening and isolation of heavy metal tolerant bacteria in industrial effluent. *Procedia Environmental Sciences*, 30, 33–37.
- Poopal, A. C., & Laxman, R. S. (2009). Studies on biological reduction of chromate by *Streptomyces griseus*. *Journal of Hazardous Materials*, 169, 539–545.
- Prokhorova, A., Stumm-Richter, K., Doetsch, A., & Gescher, J. (2017). Resilience, dynamics and interactions within a multi-species exoelectrogenic model biofilm community. *Applied Environmental Microbiology*, 83(6), e03033–e03016.

- Richter, K., Schicklberger, M., & Gescher, J. (2012). Dissimilatory reduction of extracellular electron acceptors in anaerobic respiration. *Applied Environmental Microbiology*, 78(4), 913–921.
- Roden, E. E., & Lovley, D. R. (1993). Dissimilatory Fe (III) reduction by the marine microorganism *Desulfuromonas acetoxidans*. *Applied Environmental Microbiology*, 59(3), 734–742.
- Rosenberg, E., DeLong, E. F., Lory, S., Stackebrandt, E., & Thompson, F. (Eds.). (2014). *The Prokaryotes. Prokaryotic Physiology and Biochemistry*. Springer-Verlag, Berlin, Heidelberg.
- Sharma, P., Singh, S. P., Parakh, S. K., & Tong, Y. W. (2022). Health hazards of hexavalent chromium (Cr (VI)) and its microbial reduction. *Bioengineered*, 13(3), 4923–4938.
- Simonte, F., Sturm, G., Gescher, J., & Sturm-Richter, K. (2017). Extracellular electron transfer and biosensors. In: Редактора (Eds.). *Advances in Biochemical Engineering / Biotechnology*. Heidelberg: Springer, Berlin.
- Sobel, Z., & Schiestl, R. H. (2012). Intracellular and extracellular factors influencing Cr(VI) and Cr(III) genotoxicity. *Environmental and Molecular Mutagenesis*, 53, 94–100.
- Sung, Y., Ritalahti, K. M., Sanford, R. A., Urbance, J. W., Flynn, S. J., Tiedje, J. M., & Löffler, F. E. (2003). Characterization of two tetrachloroethene-reducing, acetate-oxidizing anaerobic bacteria and their description as *Desulfuromonas michiganensis* sp. nov. *Applied Environmental Microbiology*, 69(5), 2964–2974.
- Teng, Y., Xu, Y., Wang, X., & Christie, P. (2019). Function of biohydrogen metabolism and related microbial communities in environmental bioremediation. *Frontiers in Microbiology*, 10, 106.
- Vasylyv, O. M., Maslovska, O. D., Hnatysh, S. O., Bilyy, O. I., & Ferensovych, Y. P. (2016). Electric current generation by *Desulfuromonas acetoxidans* IMV B-7384 while application of ferric citrate, fuchsine and methylene blue. *Microbiology and Biotechnology*, 4(36), 42–49.
- Vasylyv, O. M., Maslovska, O. D., Ferensovych, Y. P., Bilyy, O. I., & Hnatysh, S. O. (2015). Interconnection between tricarboxylic acid cycle and energy generation in microbial fuel cell performed by *Desulfuromonas acetoxidans* IMV B-7384. *Proceedings of SPIE*, 9493, 1–7.
- Vasylyv, O., Bilyy, O., Hnatysh, S., Kushkevych, I., & Getman, V. (2011). The changes of spectroscopic characteristics of sulfur reducing bacteria *Desulfuromonas acetoxidans* under the influence of different metal ions. *Proceedings of SPIE*, 8152, 1–7.
- Viti, C., Marchi, E., Decorosi, F., & Giovannetti, L. (2014). Molecular mechanisms of Cr(VI) resistance in bacteria and fungi. *FEMS Microbiology Reviews*, 38(4), 633–659.