UDC 579.222

V. Govoruha
O. Tashyrev,
Doctor of Technical Sciences,
Institute of Microbiology and Virology D.K. Zabolotny
National Academy of Sciences of Ukraine

Dynamics of interacting of association spore-former soil microorganisms with Fe (III)

Goal. Investigate the dynamics of the interaction of the association of spore-forming soil microorganisms with Fe (III) compounds. Methods. Microbiological, potentiometric, colorimetric methods and gas chromatography were applied. Results The dynamics of the interaction of the association of spore-forming soil microorganisms with Fe (III) compounds was investigated. It has been established that the natural microbial group maintains the stability of the function of introducing Fe (III) compounds. It was found that the efficiency of reduction of Fe (III) in 74 years was 65%, and precipitation efficiency - 35%. Conclusions The ability of an association to interact with Fe compounds indicates its significant role in the biogeochemical cycles of the transformation of Fe in the genus. The assessment of the effectiveness of the interaction of the natural soil association with Fe compounds creates the basis for the development of effective biotechnologies for purifying water from Fe, as well as the removal of its compounds from depleted deposits.

Key words: thermodynamic forecasting, biogeochemical cycles of Fe transformation, association of spore-forming soil microorganisms.

In the earth's crust, Fe compounds are the most common among heavy metals. Fe (II) and Fe (III) compounds are part of insoluble minerals: hematite (Fe2O3), magnetite (Fe3O4), goethite (α-FeO (OH)), lepidocrite (γ-FeO (OH)), pyrite FeS2), siderite (FeCO3), and others. [2, 3]. In the process of their destruction, Fe is mobilized and involved in the biological cycle [1]. Fe compounds are present in all soils, marine and freshwater ecosystems [7, 10, 11].

Thermodynamic calculations of the interaction of microorganisms with Fe compounds [8] show that the microbial transformation of Fe (III) and Fe (II) compounds has a significant effect on the redistribution of vector flows of carbon and Fe in nature.

The purpose of the work is to investigate the dynamics of the association of spore-forming soil microorganisms with Fe (III) compounds.

Spores of microorganisms are very common in the soil. Their livelihoods can significantly affect the redistribution of vector flows C (carbon) and Fe in ecosystems.

Investigation of the interaction of the spore-forming association Soil microorganisms with Fe compounds give an opportunity to evaluate their influence on the biogeochemical cycles of Fe transformation in nature.

In addition, the use of spore-forming microorganisms in the study of interaction with Fe compounds is promising from the point of view of biotechnology. Benefits of soil spore-forming association of microorganisms: common in nature; the simplicity and ease of obtaining such an association; possibility of long-term storage; a wide range of substrates; high speed of their digestion, etc.

That is why the association of spore-forming soil microorganisms was chosen as a model for studying the laws of interaction with Fe compounds [5].

The model substrate was potato, since starch is one of the most commonly used polymers in nature [5, 12]. Its destruction by soil microorganisms creates conditions for the effective restoration of Fe (III) to Fe (II) compounds.

Materials and methods. Objects of research were spore-based microorganisms, Fe (III) and Fe (II) compounds, as well as potatoes as a model substrate (natural polymer).

To obtain the association of spore-forming soil microorganisms, 50 g of potatoes cut into cubes (0.5-1 cm in length) were transferred to a 250 ml vial and filled with 120 ml of tap water. To select spore bacteria, the

vial was boiled in a water bath for 10 minutes under a cotton-marl stopper. After cooling, the bottle was sealed with a rubber stopper and a metal clamping device. The cultivation was carried out for 7 days at a temperature of 28 ° C. The development of microorganisms was indicated by turbidity of the environment, potato destruction, active foam formation, oxygen concentration reduction, hydrogen and carbon dioxide synthesis, changes in pH and Eh.

To investigate the dynamics of the interaction of bacteria with Fe (III) compounds, the resulting association of spore-forming soil microorganisms was used. The cultivation was carried out in a plastic cultivator measuring 9 × 15 × 10 cm, which was tightly sealed. The cultivator is equipped with two fittings: for selecting samples of the gas phase and the culture fluid. The sterilized cultivator was washed with 35% hydrogen peroxide solution. Sliced potatoes (50 g) were filled with 500 ml of tap water and boiled in a glass vial under a cotton gauze plug in a water bath for 10 minutes. Heat-treated potatoes and water were transferred to a cultivator and added 2 ml of a culture fluid containing spore-forming soil association. The cultivation was carried out at 28 ° C. One day after its beginning, after reduction of the redox potential to negative values and the beginning of the synthesis of hydrogen, the Fe (III) citrate was added to the vial at a concentration of 0.25 g / L by iron cation.

The following metabolic parameters were monitored: pH and Eh, hydrogen concentration, Fe (III) and Fe (II). The pH and Eh indices were measured by a thin-watt metric method. The pH meter-milivoltmeter "pH-150 MA" with the measuring electrode ESK-10603/4 was used to determine the pH. The redox potential was measured using a pH-meter-millivoltmeter "pH-150 MA" and a measuring electrode EPB-1. Comparison electrode - chlorine silver electrode EVL-1M3.

The concentration of hydrogen was determined according to the standard method on the gas chromatography LHM-8-MD [4] equipped with two steel columns - one (I) for the analysis of H2, O2, N2 and CH4, the second (II) - for the analysis of CO2. Parameters of columns: I - l = 3 m, d = 3 mm, with molecular sieve 13X (NaX); II - l = 2 m, d = 3 mm, with porapak-Q carrier; the temperature of the columns - + 60 ° C, the evaporator - + 75 ° C, the detector - + 60 ° C, the current of the detector - 50 mA. The gas carrier is argon, the gas duct velocity is 30 cm3 / min. Volume of gas samples: on column I - 2,5 cm3, II - 1 cm3.

The H2 content in the gas mixture (%) was calculated according to the standard method in terms of the area of the peaks of the components of the gas phase.

The concentration of Fe (III) and Fe (II) was determined by the colorimetric method [6]. Prior to determining Fe content, 6 ml of culture fluid was centrifuged at 2655 g for 15 min. The supernatant was poured out and used to determine the concentration of Fe compounds.

Determination of the concentration of Fe (II) is based on the formation of reddish colored Fe (II) compounds with o-phenanthroline. To measure, 1.5 ml of supernatant was taken and 0.75 ml of a 0.25% solution of o-phenanthroline was added. The presence of Fe (II) was evidenced by the appearance of a red-orange color. The amount of Fe (II) was determined using a photocoupler KFK-2MP at λ = 490 nm and an optical path length of 0.5 cm.

The concentration of Fe (III) was determined by the method based on the formation of colored Fe (III) with potassium rhodanide in acidic media. To this end, 1.5 ml of supernatant was taken, 0.25 ml of a 1.5 M solution of KSCN and 0.75 ml of concentrated HCl were added. The presence of Fe (III) was evidenced by the appearance of a red color. The amount of Fe (III) was determined using a photoelektocolorimeter KFK-2MP at λ = 490 nm and an optical path length of 1 cm.

Research results. The dynamics of the association of spore-forming soil microorganisms with Fe compounds was studied at an iron cation concentration of 0.25 g / l.

An important characteristic of the microbial association is the ability to maintain a stable functioning of the introduction of Fe (III) in high concentrations.

At the time of the introduction, the Association actively fermented the potatoes (Fig. 1). This was evidenced by a decrease in the pH of the nutrient medium from 7 to 5, and Eh - from +380 up to - 120 mV.

10 minutes after the introduction of Fe (III) redox, the potential of the culture fluid was increased from -20 to +20 mV and fluctuated within 2 hours in the range from -7 to +20 mV. Over the next 2 hours, the Eh dropped sharply to-140 mV. In the further 70 years, the redox potential decreased to-180 mV. That is, within 4 years the association adapted to the Fe introduced and restored the output of the redox potential.

The pH after the introduction of Fe (III) increased from 5 to 5.1 and subsequently remained within the range of 5.1 - 5.2.

The removal of Fe (III) did not inhibit the synthesis of hydrogen by association (Fig. 2). The concentration of synthesized hydrogen at the time of iron introduction was 31.5%. After 1 year, the concentration of H2 increased by 5%, and subsequently remained within the limits of 33 - 35%. After 74 years of cultivation, the final concentration of hydrogen was 38.5%.

The obtained results indicate the association ability to maintain the stability of the function of introducing Fe (III) at a concentration of 0.25 g / l. The rate of recovery of baseline metabolic rates, in particular Eh, suggests that the concentration of Fe (III) studied is not lethal for microorganisms and can be used to simulate their interaction with Fe compounds. Low redox potential, lack of oxygen and the presence of hydrogen in the gas phase by thermodynamic calculations [8] should contribute to the rapid and effective reduction of Fe (III) to Fe (II). The recovery of Fe (III) to Fe (II) began immediately after the introduction of Fe (Fig. 3). After 10 minutes, 0.5% of the introduced Fe (III) was restored. For 74 years, the efficiency of Fe (III) reduction by microorganisms was 65%. Reduction of the concentration of Fe (III) and the accumulation of Fe (II) in the culture liquid is not arose in the stoichiometric relationship. Therefore, we assume that the restoration of Fe (III) by microorganisms was non-specific due to the accumulation of exombetabolites, reduction of the redox potential, as well as the functioning of low-potential redox enzymes, as evidenced by the active synthesis of hydrogen.

The final total concentration of dissolved Fe (III) and Fe (II) compounds was 0.19 g / L. The remaining Fe compounds could be immobilized by microorganisms in the form of insoluble Fe hydroxides or sorbed with biomass. The efficacy of precipitation of Fe (III) and Fe (II) compounds was 35%.

The high efficiency of the non-specific reduction of Fe (III) to Fe (II), which is not adapted to Fe by the association of spore-forming soil microorganisms, the effectiveness of precipitation of Fe, a significant proliferation of such microbial natural groups, as well as the distribution of Fe compounds in ecosystems at concentrations up to several grams per 1 kg of soil [9], indicate that such microorganisms have a significant effect on the redistribution of vector flows of carbon and iron in nature.

The ability of the association of spore-forming soil microorganisms to quickly adapt to the increase in the concentration of Fe (III) and to interact with it, restoring or sediment, creates the basis for the development of effective biotechnologies for water purification from Fe compounds and increasing the efficiency of its removal from impoverished deposits.

Conclusions

The dynamics of the interaction of the association of spore-forming soil microorganisms with Fe compounds has been investigated. The ability of the microbial group to maintain the stability of the functioning of introducing Fe (III) at a concentration of 0.25 g / L and to interact with Fe compounds is established. It is believed that such an association with the compounds of Fe (III) and Fe (II) can significantly affect the biogeochemical cycles of the transformation of Fe in nature. The obtained laws of the interaction of the spore-forming soil association with Fe compounds can become the basis for the development of effective biotechnologies for purifying water from Fe (III) and Fe (II) and increasing the efficiency of extraction of Fe from impoverished deposits.

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