Groundwater microflora of the Aptian-Cenomanian deposits at the Igolsko-Talovoe field in Tomsk Region

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Abstract The authors have studied the microbiological composition of the groundwater of the Aptian-Cenomanian deposits in the territory of the Igolsko-Talovoe field in Tomsk Region. The detected diversity of the physiological groups of bacteria can be a corrosive component for waters used in the reservoir pressure maintenance system. The research findings have allowed making conclusions about the need to study the contribution of all microorganisms inhabiting the waters of the Aptian-Cenomanian deposits to corrosion.

1. Introduction

The groundwater of the Aptian-Cenomanian deposits in the territory of the Igolsko-Talovoe field in Tomsk Region has an original chemical composition and is used in the reservoir pressure maintenance system, but is characterized by enhanced corrosiveness that is traditionally connected with the activity of sulfate-reducing bacteria. However, the basis of microbial communities in the groundwater under study is formed by saprophytic, oligotrophic and nitrate-reducing bacteria. Sulfate-reducing microorganisms are present in these waters in small amounts and are characterized by low metabolic activity. Along with that, nitrate-reducing bacteria that cover the same environmental niche with sulfate-reducing bacteria are available here in considerable amounts and are able to take part in corrosive processes. Therefore, there is a need to study the corrosive activity of all groups of microorganisms living in the groundwater of the Aptian-Cenomanian deposits in order to develop effective means of suppressing corrosive processes.

2. Materials and methods

2.1. Subject of research

The administrative location of the Igolsko-Talovoe field is Kargasoksky District of Tomsk Region, its southwestern part (Figure 1). It was discovered in 1977 and has been developed by VasyuganNeft. The settlements being closest to the field are such towns as Nefteyugansk, Nizhnevartovsk and Pytyakh, as well as Maysk stationary village located at the distance of 25 km to the northeast of the field. Igol village has been constructed in the area of the field.

As for the regional structural and tectonic characteristics of the oil field, it is located in the southern part of the Nyurol depression. It is structurally confined to the Igolsko-Talovoe local elevation being an element of the big Igolskaya dome-like structure and having the central position in it.

The cross section of the Igolsko-Talovoe field is formed by the deposits of the pre-Jurassic foundation that is uncomfortably covered by the deposits of a thick sedimentary mantle. The productivity of the Igolsko-Talovaya area is connected with the regionally oil-bearing J1 horizon of the
The Vasyugan Formation. The Igolsko-Talovoe field has several big oil pools. The groundwater of the Aptian-Cenomanian deposits is very deep and covered by thick regional impermeable beds that reliably protect it against pollution from the surface [1–4].

The clusters of water wells used in the reservoir pressure maintenance system are located on both banks of the Chertala River at the distance of about 3.5 km (cluster No.9) and 4.5 km (cluster No.36) from the river bed. The complex of facilities using the groundwater of the Aptian-Cenomanian deposits is part of the pressure maintenance system of this oil pool. The areas of the water extraction installations are combined with the oil production facilities of the Igolsky oil field area.

2.2. Sampling and research methods
For research purposes, water samples were taken from production and injection wells once a quarter. Sampling was performed at the water wells from the sampler (valve) after water had been drained for several minutes. Water samples for the microbiological analysis were taken into sterile glass bottles while the rules of asepsis were observed. In the waters under consideration, the authors were determining a wide range of organotrophic microorganisms that can have this or that impact on the chemical composition of the waters in this aquifer system and oil quality, as well as on the technical condition of oil production equipment.

Identification of and quantitative accounting of microorganisms was carried out in accordance with classical techniques common in Microbiology, which are outlined in some of our publications [4–7]. Simultaneously, the authors were examining the chemical composition of the waters as a habitat of the microorganisms.

3. Hydrochemical characteristic
3.1 Microcomponents and pH
The Aptian-Cenomanian aquifer system of the Igolsko-Talovoe field is characterized by horizontal hydrogeochemical zoning being typical of the region in general. First of all, it means enhanced mineralization, presence of macro- and microcomponents in the northwestern direction, progressively with layer deepening.
Table 1. Components of groundwater chemical composition in the Aptian-Cenomanian deposits of the Igolsko-Talovoe field.

| Indicators and components | Cluster and well No. | Cluster 9, well 1c | Cluster 9, well 2c | Cluster 9, well 3c |
|---------------------------|----------------------|--------------------|--------------------|--------------------|
| Sampling interval, m      | 985-1,290            | 1,130-1,581        | 1,430-1,757        |
| Exit temperature, °C      | 48                   | 39                 | 39                 |
| pH                        | 6.8                  | 7.0                | 7.1                |
| Ca²⁺, mg/dm³              | 674                  | 662                | 687                |
| Mg²⁺, mg/dm³              | 121                  | 117                | 121                |
| K⁺, mg/dm³                | 37                   | 38                 | 37                 |
| NH₄⁺, mg/dm³              | 55.0                 | 23.0               | 67.0               |
| NO₂⁻, mg/dm³              | <0.05                | <0.05              | <0.05              |
| NO₃⁻, mg/dm³              | 0.1                  | 0.1                | 0.1                |
| CO₃²⁻, mg/dm³             | <0.10                | <0.10              | <0.10              |
| HCO₃⁻, mg/dm³             | 183                  | 159                | 207                |
| Cl⁻, mg/dm³               | 12,723               | 12,723             | 12,723             |
| Na+, mg/dm³/              | -                    | 5,000              | -                  |
| Fe, mg/dm³                | 4.1                  | 6.3                | 4.8                |
| Si, mg/dm³                | 0.10                 | 0.31               | 0.31               |
| Mineralization, g/dm³     | 20.93                | 20.92              | 20.95              |

The groundwater in the aquifer system under study at the Igolsko-Talovoe field has somewhat enhanced mineralization vs. other oil fields of Tomsk Region (except the Olenye field). Its mineralization has been staying within a narrow interval from 20.31 to 23.64 g/dm³ over the many-year period of water production from the Aptian-Cenomanian deposits of the Igolsko-Talovoe field. Its pH value is neutral (pH 6.8-7.2), sometimes weakly alkaline (7.30-7.77). The content of chloride ion is 97.7-99.3, sodium ion – 77.4-98.3 %-eq. The content of sulfate ion is low (0-50 mg/dm³), which is typical of the whole section of the lower hydrogeological stage of the West Siberian Basin. The prevailing type of waters is chloride sodium [1]. The proportion of these components and the type of water coincide with the waters of productive Jurassic deposits. The waters of this aquifer system contain a wide range of microcomponents: iodine, bromine, boron, rubidium, manganese, gold, etc. The composition of the water-dissolved gases is predominantly methane-based (Table 2). Apart from methane, they are also characterized by the significant amounts of nitrogen. They also have a small content of carbon dioxide, oxygen and ethane.

Table 2. Component composition of water-dissolved gas.

| No. | Components          | Unit of measure | Content |
|-----|---------------------|-----------------|---------|
| 1.  | Gas density         | kg/m³           | 0.685   |
| 2.  | Oxygen (O₂)         | vol.%           | 0.341   |
| 3.  | Nitrogen (N₂)       | vol. %          | 3.222   |
| 4.  | Carbon dioxide (CO₂)| vol. %          | 0.762   |
| 5.  | Methane (CH₄)       | vol. %          | 95.662  |
| 6.  | Ethane (C₂H₆)       | vol. %          | 0.010   |

The content of dissolved oxygen in the waters under study varies from 0.10 to 0.31 mg/dm³, which does not exceed the permissible level of 0.5mg/dm³. Dissolved hydrogen sulfide has not been found in the waters.
4 Results and discussion

4.1 Microbiological composition of waters

The total number of microbes in the water of the wells perforating the aquifer of the Cenomanian deposits at different depths amounts for one to 2.5 million cells/ml and grows with depth (Table 3). At the same time, the number of living bacteria drops with depth. Viable cells comprise somewhat more than a half of the total amount in the deepest well. It means that the aquifer has factors being unfavorable for the activity of microbes.

Table 3. Groundwater microflora in the Aptian-Cenomanian deposits of the Igolsko-Talovoe oil field.

| Physiological groups of microorganisms | Cluster 9, well 1c | Cluster 9, well 2c | Cluster 9, well 3c |
|--------------------------------------|-------------------|-------------------|-------------------|
| Total number of bacteria, cells/ml  | 1,120,000         | 1,690,000         | 2,440,000         |
| Number of living cells, %            | 84                | 76                | 56                |
| Saprophytic, cells/ml/%              | 200,000/21.2      | 420,000/32.8      | 80/0.0001         |
| Oligotrophic, cells/ml/%             | 57,500/6.4        | 317,200/24.8      | 600,000/44        |
| Heterotrophic iron-oxidizing bacteria, cells/ml | 660/0.05 | 870/0.05 | 12,000/0.8 |
| Oil-oxidizing, cells/ml/%            | 4,200/0.45        | 3,000/0.23        | 0                 |
| Pentane-oxidizing, standard units    | 300               | 300               | 290               |
| Hexane-oxidizing, standard units     | 350               | 390               | 390               |
| Heptane-oxidizing, standard units    | 290               | 300               | 320               |
| Octane-oxidizing, standard units     | 270               | 350               | 300               |
| Nonane-oxidizing, standard units     | 440               | 440               | 350               |
| Decane-oxidizing, standard units     | 400               | 250               | 270               |
| Benzenec-oxidizing, standard units   | 450               | 450               | 400               |
| Toluene-oxidizing, standard units    | 420               | 450               | 400               |
| Phenol-oxidizing, standard units     | 300               | 320               | 290               |
| Naphthalene-oxidizing, standard units| 360               | 490               | 430               |
| Nitrate-reducing, cells/ml/%         | 11.5*10^4/10      | 9.5*10^7/7.3      | 1.5*10^9/11      |
| Nitrifying                           | 0                 | 0                 | 0                 |
| Sulfate-reducing, cells/ml/points    | 0                 | 10/6              | 10/6              |
| Thionic, cells/ml                    | 0                 | 10                | 100               |

Viable cells form microbial communities of a specific composition being typical of each sampling interval. Despite the fact that the first two wells are characterized by the prevalence of saprophytes, their amount makes only 21 or 33 % of the living cells, whereas they are practically absent in well 3c.

The titer of oligotrophic microorganisms grows with depth. They prevail over saprophytes and make 44 % of the total number of living cells in the water of the deepest well. The titer of oil-oxidizing bacteria drastically drops with depth to make from several thousand to single cells per 1 ml. However, at all depths there is the intensive oxidation of individual hydrocarbons being part of the oil composition, even where oil-oxidizing microbes are practically absent. This fact is testified to by the presence and activity of bacteria oxidizing different hydrocarbons. Apparently, hydrocarbons and oil are oxidized in this aquifer with the help of bacteria from all physiological groups and having an adaptive inducible fermentative system [2]. The water of all wells contains heterotrophic iron-oxidizing bacteria that deposit iron hydroxide on the surface of cells. Their titer pronouncedly grows with depth. Nitrate-reducing bacteria (N*10^5 cells/ml) are found ubiquitously in significant amounts (7-11 % of the total number of living cells). Sulfate-reducing bacteria are present in the aquifer as single cells with low metabolic activity. Thionic bacteria have a very low titer. Nitrifying bacteria have not been found.

Thus, the physiological groups of bacteria in the Aptian-Cenomanian water make from 38 to 65 % of the total number of living cells. As for obvious factors, the quantitative content of specific groups of
microbes in the aquifer is influenced by the depth of a sampling interval. The total number of microbes in the aquifer grows with depth, whereas the amount of viable cells drops. These quantitative parameters characterize the microbial community of the aquifer under study as oligotrophic. Oligotrophic microorganisms are present in water from all wells, and they are practically the only inhabitants of the deepest well.

There are two problems connected with microbiologic activity for the aquifer under study: enhanced content of mechanical particles in water and their enhanced corrosive activity.

The content of mechanical impurities (particles content) in the waters under study amounts to 8.4 to 10.1 mg/dm$^3$ at the permissible levels of up to 3 mg/dm$^3$. It is found that the composition of these mechanical impurities comprises iron sulfide and oxygenated iron, clay particles, quartz, carbonates produced from the formation. Over 50% of these particles are up to 20 µm in size. A significant part of the mechanical impurities is formed by the fragments of biofilms and mineral-microbial conglomerates.

Despite the fact that sulfate-reducing bacteria in the waters under study are found occasionally, have a low titer (1-10 cells/ml), whereas the main product of metabolism, hydrogen sulfide, is present in these waters in small concentrations (0.1 to 0.5 mm/year), the speed of corrosion amounting to 0.09-0.45 g/m$^2$.

The discrepancy between the small number and activity of sulfate-reducing bacteria and corrosive activity of the waters in the Aptian-Cenomanian deposits can be reasoned by the participation of metabolites of microorganisms from other physiological groups inhabiting these waters in the corrosive process.

In particular, all samples from the waters under study are characterized by the presence of nitrate-reducing bacteria amounting to $10^2$-$10^3$ cells/ml. Nitrate- and sulfate-reducing bacteria cover the same environmental niche in rock formations [2, 3]. Both of them can successfully live and grow both in wells and in the aquifer, if they have enough sources of carbon in the form of short-chain fatty acids. There is little information on the participation of nitrate-reducing bacteria in metal corrosion. However, it is proved that the corrosive process accelerates in the presence of denitrifying microorganisms, which is connected with the generation of aggressive corrosive products of metabolism: carbon dioxide and ammonia that can lead to electrochemical metal corrosion [3].

To determine the role of nitrate- and sulfate-reducing bacteria of the Aptian-Cenomanian waters at the Igolsko-Talovoe field in metal corrosion, the authors have made an experiment with steel samples No.3 [3]. The corrosive activity of water and enrichment cultures of microorganisms was tested in compliance with GOST 9.905-82 “Corrosion test methods” and RD 39-3-973–83 “Methods of controlling microbiological contamination of oil field water and evaluating the protective and bactericidal action of reagents.” The study implied the use of flat samples made of steel No.3: three samples per test. The value of sample surface roughness was up to 6.3 µm. The weight of the samples under test varied from 7,420 to 800 mg. The samples were suspended in laboratory flasks on a fishing line. To test the corrosive activity of nitrate-reducing bacteria, the flasks were filled in with an enrichment culture with the Giltay medium and the bacteria titer of $10^3$ cells/ml. To test the corrosive activity of sulfate-reducing bacteria, the samples were doped with an enrichment culture with the Postgate medium and the titer of sulfate-reducing bacteria amounting to $10^3$ cells/ml. The experimental flasks were placed to a heated magnetic mixer. The test temperature was 20-22 °C. The test time made 6 hours. To determine the weight loss of the samples, their surfaces were cleaned. The film made of corrosion products was removed with a solution not interacting with the main metal, washed with water and defatted. Then the samples were dried and weighted. The speed of corrosion was determined with a gravimetric method on the basis of the difference between the initial weight of a sample and its weight after the test. Metal samples in the abacterial Giltay and Postgate media were used for control purposes. Weight losses were converted into the losses of units of area per annum. The values of weight loss for the samples differed insignificantly in the fourth sign and, on average, amounted to 0.002 mg for sulfate-reducing bacteria and 0.0025 mg for nitrate-reducing bacteria over
the six-hour period. On conversion to the depth of penetration, the possible corrosive activity of the microflora under given conditions will amount to 0.15-0.2 mm annually. These results characterize the corrosive activity of both types of bacteria as enhanced. Thus, the corrosive activity of nitrate-reducing bacteria is comparable with that of sulfate-reducing bacteria. Corrosion was practically absent in the control experiments without bacteria. It is necessary to take the laboratory conditions of the test into account when interpreting the results of this experiment.

Apparently, the participants of corrosive processes in the Aptian-Cenomanian waters can be microbes from all groups that are present here. Further research is needed to determine the degree of their participation in corrosive processes. This problem is rather relevant now, as corrosion-related losses are rather significant. To suppress bacterial corrosion, specialists use a wide range of bactericides, but they are not always effective enough and there are no multipurpose agents.

5. Conclusions
The present study allows concluding that the total number of microorganisms in the aquifer system of the Aptian-Cenomanian deposits in the territory of the Igolsko-Talovoe field is from one to 2.5 million cells/ml and grows with depth. Living cells comprise 56-84 % of the total number and their amount drops with depth. The composition of the microbial communities differs in the number of microbes and the diversity of physiological groups. The prevalent type in the composition of the microbial community of this aquifer system is oligotrophic microorganisms. Sulfate-reducing microorganisms in the waters under study are found in insignificant amounts and have low functional activity. The enhanced corrosive activity of the waters in this aquifer system might be reasoned by the presence of nitrate-reducing bacteria that cover the same environmental niche with sulfate-reducing bacteria. Following the quantitative accounting results, it is possible to conclude that saprophytic, oligotrophic, iron-oxidizing and oil-oxidizing microorganisms in the composition of the microflora under study can have an impact on corrosive processes.

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