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Groundwater chemical and microbiological composition in Aptian-Cenomanian deposits (Kaimisovsky oil-gas bearing province)

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Abstract. This paper reveals the investigation results of the groundwater chemical and microbiological composition in Aptian-Cenomanian deposits, Kaimisovsky oil-gas bearing province. The mineral-forming behavior of the groundwater was evaluated. It was determined that the diversity of microbial communities could be corrosive- harmful for the groundwater used in reservoir pressure maintenance systems. According to the research results it is necessary to study the groundwater microorganisms in Aptian-Cenomanian deposits and their influence on groundwater itself.

1. Introduction
Water-flooding is the major method in reservoir pressure maintenance during oil field development. Geochemical processes interacting with layer-injection water co-operation result in emerging produced water, saturated inorganic salts, salt precipitation in the wells, and sometimes further suppressing corrosive processes [1-5].
Kaimisovsky oil-gas bearing area is located within the southern West-Siberia mega-province, territory of 140 thousand km^2. This area includes Kaimisovsky and Mezhovky arch, Nurol and partially Ugansky depressions, Upper-Demyanosky megaswell and others. Within this province there are about 50 oil fields confined to anticline and / or combined reservoirs. Kaimisovsky oil-bearing province embraces approximately 2.3% of the hydrocarbon reserves in West Siberian mega-province [6]. Aptian-Cenomanian aquifer system within Kaimisovsky oil-bearing province was intensively developed for water production and further usage in the reservoir pressure maintenance system over a number of years (fig. 1).
The authors studied the hyrogeochemical and microbiological processes, as well as sediments formed from fresh groundwater [7,8]. This research is a continuation of previous research investigations. The research target object is saline groundwater of Aptian-Cenomanian aquifer system within Kaimisovsky oil-bearing province itself.

2. Materials and methods
Water samples for microbiological analysis are collected in sterilized glass bottles in accordance with aseptic regulations. A wide spectrum of microorganisms affecting both the water chemical composition in investigated aquifer system and oil quality and technological petroleum production equipment service were identified. Simultaneously, the chemical composition of water, as well as water as an environmental niche for microorganism was also investigated.

Figure 1. Overview map of Kaimisovsky oil-gas bearing province.

Identification and quantitative estimation of microorganisms are determined on the basis of traditional methods. The aerobiotic and anaerobic bacteria involved in the carbon, nitrogen and sulfur cycle were detected and calculated in the water samples from production wells. The studied spectrum of physiological bacteria group included: saprophile, oligotrophic, oil oxidizing microorganisms, autotrophic, thionic, ammonifying, dentrification, nitrite, sulfur reducing, as well as oxidizing hydrocarbons of methane group, aromatics, saturated and unsaturated bacteria. The traditional methods based on elective habitat are used in investigating the groundwater microflora [11]. Investigated water plates on solid medium (Petri dish) and on fluid medium (test tubes). The plate number of grown colonies was calculated. Limiting dilution culture was used for plating in vitro. The number of bacteria is determined by visual environment change, typical for each physiological bacteria group. Growth intensity was evaluated in scores and conditional units. MacCready tables, based on statistic methods, were used in the quantitative estimation of growths. Saprophile are cultivated on meat-and-peptone agar at temperature 20–22 °C. Grown colonies were calculated two days after seeding (plating). Oil oxidizing microorganisms are used in the metabolism of petroleum and its derivatives, which were revealed in agarized Muntz medium with oil as the only source of carbon and energy. The seeding was cultivated at 20– 22 °C during two weeks. At the end of inoculation the number of colony growths was calculated. The quality control involved the seeding from one and the same water tube in solid Muntz medium without oil. Seeding was performed four-times. Bacteria oxidizing individual oil hydrocarbons were revealed in fluid Muntz medium. Seeding was conducted in aseptic serum vials placed in disiccator at the atmosphere comparable to hydrocarbon atmosphere three times. Inoculation period was 2 weeks, after which the growth of the microbes in the tubes was evaluated according to the forming films based on conditional unit in Mogilevski. Nitrite bacteria were revealed in the medium by Vinogradsk method of limiting dilution. Seeding inoculation period was for two weeks at 22 °C. The appearance of nitrites and nitrates in the medium are evidence of the desired bacteria group. Dentrification bacteria were revealed in the medium by Giltaya method of limiting dilution during 2-week inoculation at 22 °C. The occurrence of bacteria in the medium is the result of pH changes, color of cultured liquid, nitrogen and /or ammonia which could be evident in the smell and foam. Oligotrophic bacteria were revealed in agarized distilled water without organic substances. Water plates (0.1 or 0.05ml) on Petri dish, whereas seeding was cultivated at room temperature during 2-3 weeks. Colony growths were calculated by increasing through colonometer. Sulfur-reducing bacteria were cultivated in the Postgate medium with calcium lactate as source of organic substances. Seeding was conducted by limiting dilution. Intensive sulphate reduction could be observed due to the black iron sulphide sediment in the medium formed as a result of hydrogen sulphide reaction (microbic sulphate reduction) and existing iron in the medium. Psychrophile (20 °C) and thermophile (45 °C) types of sulphate-reduction were revealed.

3. Results and Discussion

3.1. Chemical composition of waters

Aptian-Cenomanian aquifer system embraces horizontal hydrogeochemical zonation, typical for this region. This involves increase of mineralization, content of macro- and micro-components north-westward. Mineralization varies from 18.03 (Pervo-Maiskoe field) to 21.18 gr/l (Katilginskoe field); however, throughout most of the territory mineralization includes 20gr/l. Predominate water type is chlorite nitrate. Basically, aquifer water exhibits weakly acid reaction (pH ranges within 7.0-7.6), rather low content and/or no sulphate -ions. Water in Aptian-Cenomanian aquifer system embraces a wide spectrum of micro-components (table 1).

Table 1. Groundwater chemical component composition content in Aptian-Cenomanian deposit within studied areas (based on averaged data).

Components	Deposits				
	Pervo-Maiskoe	Krapivinskoe	Dvurechenskoe	Zapadno-Moiseevskoe	Lineinoe
M,mg/l	18.03	19.21	19.20	19.37	20.9
pH	7.1	7.3	7.6	7.5	7.3
Ca^{2+}	514	720	680	635	662
Mg^{2+}	136	403	329	357	117
Cl^-	10956	11998	11491	11780	12723
$Fe_{общ}$	2.5	1.5	1.5	1.5	6/3
NH_4^+	29.0	27.8	23.7	25.0	23.0
HCO_3^-	165	190	171	185	159
SO_4^{2-}	2.0	45.2	39.0	38.8	29.0
K^+	34	39	36	33	38.0
Na^+	6469	5150	7425	6778	7299
Br^-	54.3	43.7	51.6	52.1	55.8

The following conclusions are based on the investigated results: Aptian-Cenomanian aquifer groundwater is in non-equilibrium to the majority of sulphates and chlorites, but in equilibrium to carbonates (calcite, magnesium, dolomite and siderite) under the thermobaric conditions governing this complex. Carbonate minerals are also found in the sediments incrustating petroleum production equipment, especially on electrical driven centrifugal pumps.

To study the mineral-forming behavior of groundwater, physico-chemical calculations were conducted by applying software tool HydroGeo [9, 10]. Experience in the use of the last shown by us in many works. It should be noted that obtained calculations of positive equilibrium index values prove the fact that these systems could form this or that mineral, while negative values- reveal their deformation (table 2).

Table 2. Groundwater saturation degree (L) in carbonates, sulphates and chloride minerals.

Deposit	Equilibrium (L > 0)	Sub-equilibrium (0 > L > -5)	Moderately undersaturated (-5 > L> -10)	Undersaturated (-10 >L >-15)
Pervo-Maiskoe	$CaCO_3$к, $MgCO_3$, $CaMg(CO_3)_2$, $FeCO_3$	$CaCo_3$a, $SrCO_3$, $SrSO_4$	NaCl, $CaSO_4$	KCl
Krapivinskoe	$CaCO_3$к, $MgCO_3$, $CaMg(CO_3)_2$, $FeCO_3$	$CaCo_3$a, $SrCO_3$, $SrSO_4$	NaCl,$CaSO_4$	KCl
Dvurechenskoe	$CaCO_3$к, $MgCO_3$, $CaMg(CO_3)_2$, $FeCO_3$	$CaCo_3$a, $SrCO_3$, $SrSO_4$	NaCl	$MgCO_3$
Zapadno-Moiseevskoe	$CaCO_3$к, $MgCO_3$, $CaMg(CO_3)_2$, $FeCO_3$	$CaCo_3$a, $SrCO_3$	NaCl, $CaSO_4$	NaCl, $CaSO_4$
Lineinoe	$CaCO_3$к, $MgCO_3$, $CaMg(CO_3)_2$	$CaCo_3$a, $FeCO_3$	$CaSO_4$	KCl

3.2.Microbiological composition of waters

Microbiologic sampling of Aptian-Cenomanian aquifer deposit groundwater within the oil field territory of Kaimisovsky oil-bearing province was conducted for some years. These investigations showed that Aptian-Cenomanian aquifer deposit is a unique groundwater ecosystem, characterized by diverse physiological bacterial groups (table 3).

Table 3. Groundwater microflora of Aptian-Cenomanian aquifer in oil fields of Kaimisovsky oil-bearing province.

Physiological bacterial groups	**Deposits**				
	Pervo-Maiskoe	Krapivinskoe	Dvurechenskoe	Zapadno-Moiseevskoe	Lineinoe
Oligotrophile, kl/ml	159520/38	84880/79.6	450/6.7	650000/51.8	440000/46.3
Saprophile, kl/ml	256760/61	14500/13.6	5270/78.0	600000/47.8	510000/53.6
Oligotrophic index	0.2	5.0	0.0	1.0	0.0
Ammonifying, kl/ml	1000/0.23	100/0.09	100/1,5	1000/0.1	100/0.01
Nitrifying, kl/ml	0/0	0/0	0/0	0/0	0/0
Denitrifying, kl/ml	1000/0.23	0/0	200/1.5	100/0.01	1000/0.1
Hydrocarbon-oxidizing c.u.	8360/2	6050/5.7	720/10.7	3970/0.31	600/0.06
Pentanol-oxidizing, c.u.	0	300	170	300	120
Haxanlo-oxidizing, c.u.	0	310	330	250	150
Heptanon-oxidizing, c.u.	150	0	110	0	0
Octane-oxidizing, c.u.	0	150	100	0	0
Nonan-oxidizing , c.u.	0	300	180	300	200
Decane-oxidizing, c.u.	250	150	110	0	230
Beneze-oxidizing, c.u.	120	150	0	300	150
Toluol-oxidizing, c.u.	150	200	60	270	0
Phenol-oxidizing, c.u.	300	0	0	0	0
Naphtalene-oxidizing, c.u.	320	0	0	0	120
Thione-authophytic, kl/ml	1000/0.23	10/0.1	10/0.14	100/0.01	100/0.1
Sulphate-reducing, kl/ml	0/0	0/0	100/1.5	0/0	0/0

Note: left- number of cells in 1ml of water; right- number of microbes in one specific physiological group, % comparable to total microbe number in water from the deposit

The initial segment in the microbic trophic chain, degrading organic substances, including oil, is aerobiotic and anaerobic organotrophic microorganisms. Aerobiotic organotrophic microflora, such as saprophiles, oligotrophiles, ammonifying and oil-hydrocarbon oxidizing bacteria are predominate in the investigated groundwater. Saprophiles are permanent and predominate (except in very rare cases). These microorganisms are used as carbon and nitrogenous nutrition in both protein and organic compounds (including oil). During the oxidation of organic substances saprophiles form different intermediate metabolism products, especially organic acids: oxalic, citric, vinic, glutamic, fulvic and other acids which are directly and/or indirectly attack metallic equipment.

Saprophile content is one to tens of thousands kl/ml in the formation waters of some of Tomsk Oblast oil fields. In the case of deep groundwater not confined to oil fields, the number of saprophiles is cells, rarely tens in 1ml of water. Based on the investigation results of groundwater from Aptian-Cenomanian aquifer system within most of the oilfields the number of psychrophile saprophytes is hundreds kl/ml. Maximum psychrophile saprophyte content was determined in Zapadno-Moiseevskoe field groundwater (600000 kl/ml/) and minimum- Dvurechenskoe (5270 kl/ml). This fact proves that habitat factors in Aptian-Cenomanian aquifer system groundwater are favorable for life sustaining saprophile bacteria. According to the morphological forms of saprophile microflora these bacteria are predominantly sporeless rod-like bacteria. Coccus-form bacteria are rare. It is well-known that globular (spherical) bacteria are prevailing in the mature stage of organic matter degradation within the water ecosystem. Rod-like bacteria abundance indicates the increased number of hard degradable organic substances.

Oligotrophiles are widely spread, except within Dvurechenskoe oil field, where quantity distribution of saprophile bacteria is more significant. These microorganisms can exist in ecosystems with low organic substance content (0.1 – 1 mg/l). The number of oligotrophiles is a direct indicator of the mineralization level of organic substances. The microflora content in Aptian-Cenomanian aquifer system groundwater embraces less than 50% oligotrophile bacteria of all microbes, except in Krapivinskoe oil field. The quantitative margin of oligotrophiles in different oil fields ranges from thousand to hundred thousands kl/ml. Maximum number of these bacteria can be found in the groundwater within Pervo-Maiskoe oil field. The oligotrophile- saprophile ratio is the so-called oligotrophic index. If more than 1, active processes of organic substance decomposition are in progress in the ecosystem. If less than 1 mineralization of organic substances is not in progress and, as a result, ecosystems lose the capacity of self-purification. According to obtained results, microbic decomposition of organic substances occurs only in the groundwater of Krapivinskoe and Zapadno-Moiseevskoe oil fields.

Number of hydrocarbon-oxidizing bacteria in groundwater of studied oil fields is insignificant and is only from a basis point to 10% of total amount. Maximum number of bacteria in Pervo-Maiskoe oil field groundwater is 8360 kl/ml, while minimum- in Lineinoe oil field groundwater. These hydrocarbon-oxidizing bacteria have oxidation capacity towards a wide spectrum of individual hydrocarbons, i.e. not only methane group, but also aromatic group (beneze-, toluol-, phenol-, naphtalene-) (table 3). In spite of the cosmopolitan distribution, the intensive development of hydrocarbon-oxidizing bacteria is rather insignificant- 200-300 c.u. (maximum 500 c.u.). Such a diversity of hydrocarbon-oxidizing organisms is indicative of dissolved hydrocarbons in the groundwater which have migrated from the underlying oil-bearing formation of Jurassic sediments as gaseous emanation.

Aptian-Cenomanian groundwater in each of investigated oil fields embraces its own specific spectrum of hydrocarbon -oxidizing bacteria, being formed consistent to the ecosystem conditions. The role of hydrocarbon -oxidizing bacteria in investigated microbal communities involves oxidation of hydrocarbons and further formation of partially-oxidized intermediate products -spirit, aldehyde and others. Consistent composition of hydrocarbon -oxidizing bacteria includes predominately rod-like bacteria, possessing active oxidation activity of oil and its derivatives: Pseudomonas, Arthrobacter, Rhodococcus, Corynebacterium and Mycobacterium. Oil-oxidizing bacteria decomposing oil and gaseous hydrocarbons terminate the cycle by freeing carbon from the stable compounds to atmosphere as carbon dioxide.

Ammonifying bacteria – the initial chain in the nitrogen cycle within the hydrosphere, and found in all investigated oil field groundwater. However, this group is less in number comparable to the above-described ones and is only ten and hundred units kl/ml. Microbes in the next nitrogen chain- nitrifying bacteria within investigated groundwater were not detected. Thus, nitrogen cycle in Aptian-Cenomanian groundwater becomes “disconnected”. This could be associated with insignificant ammonifying bacteria productivity revealed in the investigated groundwater.

Due to oxygen consumption during hydrocarbon oxidation, oxygen deficiency emerges, promoting the development of facultative-anaerobic and anaerobic microorganisms. i.e. denitrifying and sulphate-reducing bacteria.

Denitrifying bacteria were revealed throughout all oil fields except in Krapivinskoe oil field. The number of which is tens to hundreds kl/ml. These bacteria reduce nitrates to molecular nitrogen with conjugated oxidation of organic acids, sugar and spirit to gas and water. Liberated energy (during oxidation) is used by the microorganisms for life sustainability. Denitrification could occur in aerobiotic and anaerobic conditions; however, it is more intensive in conditions without oxygen. It has been established that nitrogen source for these bacteria is nitrogen- containing heterocyclic oil components. Denitrifying bacteria are within the identical ecological niche (habitat) as sulphate-reducing bacteria, consuming both free and bound oxygen and forming favorable ecosystem (habitat) for sulphate-reducing bacteria.

Sulphate-reducing bacteria have been found only in the groundwater of Aptian-Cenomanian sediments within Dvurechenskoe oil field. Thionic bacteria were revealed in Aptian-Cenomanian groundwater within the territories of all the oil fields, the number of which is from one to tens thousand kl/ml. Maximum number of these bacteria was revealed in Pervo-Maiskoe oil field groundwater. Thionic bacteria attack forming favorable conditions for sulphate-reducing bacteria. Moreover, thionic bacteria, as other aerobiotic bacteria, consuming oxygen form favorable conditions for sulphate-reducing bacteria. However, during metabolism sulphur, pyrite and life sustaining products of sulphate-reducing bacteria oxidize into sulphides, while hydrogen sulphide- into sulphates, forming energy sources for sulphate-reducing bacteria.

4. Conclusions

Aptian-Cenomanian aquifer system within Kaimisovsky oil-bearing province has been being intensively developed for water production and further usage in the reservoir pressure maintenance system. Research results proved the significance of studying the chemical microbiological composition of above-mentioned groundwater. It is important if the water-flooded layer excludes salt-deposition and further suppressing corrosive processes. Obtained results showed that not only sulphate-reducing bacteria, but also organotrophic microflora found in groundwater, throughout its life sustaining activities could further corrosive active metabolites. In this case, this fact should be considered in oil field development.

References

[1] Frank Y, Banks D, Avakian M, Antsiferov D, Kadychagov P and Karnachuk O 2016 *Geomicrobiology Journal* Firmicutes is an important component of microbial communities in water-injected and pristine oil reservoirs, Western Siberia, Russia **Vol.** 33 (5) pp. 387-400

[2] Kashavtsev V and Mishenko I 2004 *Moscow. Orbit.* Scaling in oil extraction p 432

[3] Nalivaiko N, Dutova E and Spiridonov T 2016 *IOP Conference Series: Earth and Environmental Science.* Groundwater microflora of the Aptian-Cenomanian deposits at the Igolsko-Talovoe field in Tomsk Region **Vol.** 33 (1)

[4] Nazarov A 2004 *Idea-Press.* Oil and gas hydrogeochemistry of the South-Eastern part of the West Siberian oil and gas province p 288

[5] Trifonov N, Nazarov A and Alexeev S 2016 *IOP Conference Series: Earth and Environmental Science.* Groundwater compatibility with formation water and pay zone rocks in Pervo-Maiskoe oil-gas-condensate field to maintain formation pressure **Vol.** 33 (1)

[6] Kontorovich A, Nesterov I, Salmanov F 1975 *Nedra Press. Moscow.* Geology of oil and gas in West Siberia p 521

[7] Pokrovsky V, Dutova E, Kuzevanov K, Pokrovsky D and Nalivayko N 2015 *IOP Conf. Seri.: Earth Environ. Sci.* Hydrogeological conditions changes of Tomsk, Russia **Vol.** 27 pp. 12031

[8] Dutova E, Vologdina, Pokrovsky D, Nalivaiko N, Kuzevanov K and Pokrovsky V 2016 *IOP Conference Series: Earth and Environmental Science.* Hydrogenous mineral neoformations in Tomsk water intake facility from underground sources **Vol.** 33 (1)

[9] Dutova E, Bukaty M, Nevol'ko A, Pokrovsky D and Shvartsev S 2006 *Geologia and Geophysics.* Hydrogenic concentration of gold in alluvial placers of the Egor'evsk area (Salair) **Vol.** 47 (3) pp. 364-376

[10] Shvartsev S and Dutova E 2001 *Geology of Ore Deposits*. Hydrochemistry and mobilization of Gold in the hypergenesis zone (Kuznetsk Alatau, Russia) **Vol.** 43 (3) pp. 224-233

[11] Netrusov A, Egorova M, Zakharchuk L 2005 *Moscow. ACADEMA. Practical guide on microbiology* p 608