# Biotesting of modeled drilling mud as an indicator of environmental risk

# A S Mushunina, S V Azarova, E G Yazikov, I A Parygina

Department of Geoecology and Geochemistry, Institute of Natural Resources, National Research Tomsk Polytechnic University, 30 Lenin Av., Tomsk, 634050, Russia

E-mail: sashenbka@yandex.ru, svetazara@tpu.ru

#### Abstract.

In the system of environmental monitoring, biomonitoring plays an important and independent role. The essence of the method determining the toxicants' effect consists in the specially selected organisms under standard conditions and recording different behavioral, physiological and biochemical factors. The paper studies the biological influence of modeled drilling mud components. The research results have shown the toxic effect of investigated muds. The obtained data confirm that biotesting can be applied for drilling fluids certification by the complex assessment of hazardous substances.

#### 1. Introduction

The biotesting methods of environmental management have found wide application in the sphere of detection of pollutants hazard. Such methods are known to be widely implemented abroad by environmental monitoring and protection organizations. One of the advantages of biotesting is detection of toxic substances, most of which are not specified by the operating standards, but can lead to the toxic or mutagenic effects. Today, a wide range of test objects have been studied: from microorganisms to animals. Each of them has its own characteristics, advantages and disadvantages, knowledge of which helps to choose an indicator with appropriate "sensitivity" to definite pollutants. For example, since 1985 in the USA there has been an obligatory toxicological biotesting for drilling fluids with the use of mysids as a test object [1]. In Russia, water biotesting using daphnids and infusoria is included in the obligatory list of the National State Standard 17.1.2.04-77 for fishery waters [2].

Ready and waste drilling muds are chemical products, which are an integral part of drilling process in oil industry. For their preparation, a wide range of organic and non-organic substances, chemicals and additive compounds are used [3, 4, 5, 6]. In the USA more than 1900 different components are produced by about 100 enterprises. The used materials are sent to a sludge reservoir (a pit) after operating cycle in the well. According to some authors' data [7, 8, 9], production activity connected with the drilling muds, which are the part of drilling waste, affects negatively some environmental components. Transfer of drilling muds, drilling flush fluids and other chemical products into the environment is dangerous in spite of its self-purification capacity. Drilling cuttings and waste water contact the drilling mud and become toxic. The stored drilling mud components are toxic by themselves. Besides, it is impossible to prevent completely the potential accidents. The research goal is to assess the hazard of drilling mud components for the environment using biotesting.

The relevance and practical application of the research is defined by the opportunity to assess and to predict the biological impact of each component of certain chemical elements using modeled drilling muds. Having analysed the results obtained by means of Chlorella vulgarisbeijer weed, it is possible to simulate the effect of water-soluble substances in studied materials. The use of Drosophila melanogaster fly allows studying the effect of these components (including solids). In addition, the obtained results can be used for development of substances list in the National State Standard 17.1.2.04-77.

Toxicity detection of modeled drilling mud components is necessary due to the emission of one major ingredient used in drilling.

#### 2. Methods and materials

For the first time Chlorella vulgarisbeijer weed and Drosophila melanogaster fly are used as test objects to study the biological effect of drilling muds. The toxicity was experimentally estimated in the analytical laboratories (Tomsk Polytechnic University) following the requirements of the guidance. According to "Toxicity detection of drink, natural and waste waters, soil-water extracts, sewage sludge production and consumer waste for optical density measurement of Chlorella vulgarisbeijer weed" [10], a series of experiments were conducted (8 samples) to determine toxicity of drilling mud water extracts in the Fundamental Research Laboratory of Hydrogeochemistry (analyst: Vorobieva D.A.). The analysis includes measurement of optical density of weed suspension. The special multiple-cell propagator allows timely revealing the changes in cell number in the reference and experimental samples in acute toxicological condition. A criterion is a growth inhibition or promotion in values of weed optical density [11].

To detect content of the toxic substances sample biotesting was conducted using Drosophila melanogaster fly as a test object in the Department of Geoecology and Geochemistry according to the recommendations applied for the extractive industry waste [11], medicines, and urban dust. The results were statistically processed with the fitting criterion -X2. During biotesting 5 samples were studied at 1% concentration of basic material in the medium. For the experiment, yellow (y) and singed (sn) Drosophila lines were taken. The yellow ones had yellow bodies and straight bristles, the singed ones had grey bodies and scorched bristles. Two females and one male were placed in the test-tubes with the prepared medium for two days. Then, the development of new generation was controlled. The changes in external features and ratio of males indicated the impact of studied muds. The reference and experimental groups were formed simultaneously and identically. The lethal dose (LD<sub>50</sub>) was determined for every sample.

#### 3. Results and discussion

The research conducted in 2000 by P.S. Chubik in Tomsk Polytechnic University proved the urgency for development of environmental friendly drilling fluid. In his paper it was mentioned that the ecotoxicity assessment of drilling flush fluids should be made at all stages of their "life cycle" to obtain reliable data, which can be achieved only by instrumental biotesting [8, 12].

The authors examine the "first" life cycle of drilling muds. These are modeled drilling muds produced in the chemical laboratory of non-production cycle.

Actually, there is no universal standard composition of drilling muds. Their application depends on location. Besides, they can drastically differ in terms of a well depth and different ore structure. Currently, no special universal methods of hazard assessment of drilling muds or their compounds for the environment have been developed. The characteristics of mud cellulose esters-based components are estimated as non-hazadous.

The reagents of Russian origin (polyanionic cellulose (PAC)) and those of foreign origin (carboxymethylcellulose (CMC 85/1000)) were selected for analysis. It is generally accepted that the polyanionic cellulose reagents are not toxic.

Polymer-containing drilling muds started to be applied in the first half of the 1970s, and at the moment, they are the most commonly and widely used reagents in drilling. So, for biotesting, reagents most of which are polysaccharides were chosen [12]. Production of water-soluble cellulose ethers reaches about 380 thousands of tons per year, with 180 thousand tons of carboximetyl cellulose (CMC). The use of polymeric reagents (CMC) in mud composition has made it possible to improve the wellbore wall conditions, limit the oil content in the mud, and enhance the well cementing quality.

CMC is an imported reagent, which is an interaction product of cellulose with monochloroacetic acid. Dry sodium salt CMC has a weak corrosive action. The main property of the reagent is a gelation [13-18].

Polyanionic cellulose (PAC) is made from the same basic substances as a typical CMC. There are various modifications of the production technologies, which allow obtaining PAC with different degrees of substitution. PAC is characterized by increased bioresistance to the polymineral exposure.

"Polypac R" contributes to formation of a thin, dense, firm and tight filter cake, reducing the area of filtrate invasion into the reservoir, preventing the invasion of solids; it also reduces the risk of differential sticking. The reagent is resistant to bacterial exposure; it does not require any bactericide that complicates its further processing by the bacterial recycling.

PAC (BB, HV) is a multi-purpose reagent used as a thickener, clay inhibitor, lubricating staff, and agents to reduce the loss of drilling fluid. The substitution degree is not less than 90; Brookfield viscosity ranges from 120 to 4500. The country of origin is Russia. The type of reagent is a surface-active agent. In terms of salt resistance the reagent is 10% NaCl salt resistant. Regarding heat resistance the reagent is resistant. In terms of application the reagent is a clay rock inhibitor.

CMC. The polymerization degree of up to 1200 is used to increase viscosity of CMC drilling mud with the polymerization degree of up to 85 / 1000-85 / 1200 in drilling deep wells. The country of origin is Russia. The reagent type is a surface-active agent. In terms of salt resistance the reagent is limited 3 - 10% NaCl salt resistant. In terms of heat resistance the reagent is estimated as resistant. As for application the reagent is used as a filtrate reducer.

"Polypac R" is polyanionic cellulose of high quality, water-soluble polymer used for control of water-based muds filtration, manufacturer is MI-SWACO. The country of origin is the USA. "Polypac R" is used to reduce water loss of fresh, salted, potassium chloride and salt-saturated muds as well as sea water muds. It promotes the formation of a thin, dense, firm, and tight filter cake, reducing the area of filtrate invasion into the reservoir, preventing the invasion of solids; it also reduces the risk of differential sticking. The reagent is resistant to bacterial exposure; it does not require the use of bactericides. "Polypac R" is cost-effective and efficient.

# 4. Using Chlorella vulgarisbeijer weed as a test object

Methods of toxicological analysis using Chlorella vulgarisbeijer weed were approved for the state environmental monitoring purposes. It can be applied to certify drilling muds in combination with other test objects for the comparative evaluation of drilling mud toxicity and hazard class definition.

Using the Kichemasov's method [10], the quality of the water extract of the studied components was determined in terms of their toxicity. For this purpose, the relative difference (in %) of the optical density value for each dilution in comparison with the reference one was calculated. As a result, in all the studied samples the toxicity is indicated (table 1).

N⁰	Drilling mud component	Value of toxic multiplicity of dilutions	Frequency r*, %	Result of toxicological analysis (quality of water extract)	
1	CMC	7	10	Moderately toxic	
2	Polypac R	4.4	3	- Moderatery toxic	

Table 1. Determination	ection of water sa	nple quality in	n drilling mud	l components
------------------------	--------------------	-----------------	----------------	--------------

doi:10.1088/1755-1315/43/1/012047

IOP Conf. Series: Earth and Environmental Science 43 (2016) 012047

3	Polypac R+ NaCl	8	21	
4	PAC HV	24	12	
5	CMC +NaCl	9	5	Torio
6	PAC BB+ NaCl	22	7	
7	PAC HV+ NaCl	9.6	15	_
8	PAC BB	59	23	Highly toxic

Based on the research results, it can be concluded that the toxicity degree of studied components is increasing with the addition of salts in spite of the fact that salts are generally not toxic. Presumably, the interaction of additional components with the drilling mud base (cellulose esters) may cause increased toxic effects.

#### 4. The use of Drosophila melanogaster fly as a test object

For a long time, biotesting on Drosophila flies has been used in the medicine. Drosophila melanogaster is the most studied object, which helps to identify the effect of a substance on the living organisms. Different parameters of Drosophila melanogaster are widely used for hazard assessment of chemicals [19, 20].

Using fruit flies in the experiments with the drilling mud components, the following biological factors are estimated: sex ratio and morphoses in relation to sample concentration in the medium. The values of these factors in the experiment groups are compared with the values in the control groups. When processing the data, the statistical index X2 is used to determine the conformity degree of experimental data to expected ones. The obtained results are presented in table 2.

NG	Drilling mud component	Sex ra	Mambagag 0/		
JN⊻		Males	Females	Morphoses, %	
1	CMC	43	57	7.5	
2	Polipac R	28	72	14	
3	PAC BB	48	52	39	
4	Duo-vis	17	82	25	
5	Control	50	50	1	

Table 2. Quantitative description of studied factors.

Obtained X2 values in the components: Polypac R, Duo-vis exceed the critical value of 3.84 (p = 0.05; at the number of freedom degree equal to 1) for Polypac R. Hence, the suggestion that male/female ratio in the reference medium and in the medium with sample addition is equal (1:1), is incorrect for Duo-vis. So, it can be concluded that the studied compound samples have influenced the Drosophila flies sex ratio. In the components of PAC-BB, CMC X2 value does not exceed 3.84; it shows that there are no samples with toxic effect for this factor. The "morphoses presence" factor was calculated in a similar way. The drilling mud components Polypac R, PAC-BB, Duo-vis belong to the toxic substances. Therefore, the use of Drosophila melanogaster fly as a test object allows referring Polypac R, PAC-BB and Duo-vis to the substances with toxic effect. The summarized data on the studied test objects are shown in table 3.

Table 3. Summarized data on the studied test objects.
-

N⁰		Test objects	
	Drilling mud components	1	2
1	CMC		n.d
2	Polypac R		
3	Polypac R+ NaCl		n.d
4	PAC HV		n.d

IOP Conf. Series: Earth and Environmental Science 43 (2016) 012047

doi:10.1088/1755-1315/43/1/012047

5	CMC +NaCl		n.d	
6	PAC BB+ NaCl		n.d	
7	PAC HV+ NaCl		n.d.	
8	PAC BB			
Note: 1 - Chlorella vulgarisbeijer, 2 - Drosophila melanogaster				
– presence of biological effect				
- absence of biological effect				

*n.d.* – not detected

The analysis performed shows that toxicity was detected in all studied samples using different test objects.

# Conclusions

The research findings confirm the presence of the toxicity in drilling mud components. Chlorella vulgarisbeijer weed and Drosophilia melanogaster fly can be reliable environmental risk indicators of drilling mud components. Biotesting can be recommended for certification of drilling muds for the purpose of state environmental control.

# **Reference:**

[1] Abdullin R A 1990 *Moscow: Izd VNIIEgazprom*. New technological tools and processes providing the drilling costs reduction and environmental protection Drilling of gas and gas-condensate wells. Pp. 52.

[2] Standard 17.1.2.04-77 Protection of nature. Hydrosphere. Indicators of condition and taxation rules of fishery water bodies

[3] Heinze T, Liebert T 2001 *Progr. Polym. Sci.* Vol. **26** pp.1689.

[4] Balser K, Hoppe L, Eicher T, Wendel M and Astheimer AJ 1986, in: Ullmann's Encyclopedia of Industrial Chemistry, 5th ed W Gerhartz, YS Yamamoto, FT Champbell, R Pfefferkorn, JF Rounsaville Eds VCH Weinheim New York pp. 419.

[5] Chowdhury J K 1924, Biochem. Z 148 76.

[6] Ogawa T J 1976 Jpn. Soc. Starch Sci. Vol. 23 pp. 49.

[7] Novikov V S 1977 *Overview Drilling Series*. The results of industrial tests of potassium solution. Vol. **6** pp. 32-36.

[8] Chubik P S 2000 Methodological fundamentals of the quality optimization of drilling flush fluids: the dissertation of the doctor tehn. sciences: 05.15.14: protected 04.05.00 pp. 370.

[9] Kaufov M A, Khandokhov T X and Kerefova M K 2013 *Fundamental Researches Magazine*. Morphoses of Drosophila melanogaster by irradiation with alternating magnetic field of different frequency. Vol **10** (**10**) pp. 2219-2221.

[10] Kichemasov A N 2012 Methods Toxicity detection of drink, natural and waste waters, soilwater extracts, sewage sludge, production and consumer waste for optical density measurement of Chlorella vulgarisbeijer weed FR.1.39.2007.03223 (Moscow: Publishing House Akvaros) pp. 42.

[11] Azarova S V, Yazikov E G and Ilyinskikh N N 2004 *Tomsk, Tomsk Polytechnic University news.* Assessment of environmental risks of mining enterprises waste in the Republic of Khakassia using the biotesting method. Vol. **4** pp. 55-59.

[12] Drovnikov P G 1992 *Moscow: Publishing house VNIIOENG*. Methods for neutralization of drilling wastewater by the construction of deep wells Construction of oil and gas wells by land and by sea. Vol. **5** pp. 18-20.

[13] Stigsson, V, Kloow G and Germgard U 2001 Paper Asia pp. 16.

[14] Thewlis B H 1969 21 21 Stärke.

[15] Hsieh, P T, Chen C S and Yang C T Hua Hsueh 1978 on February 29, Chem. Abstr. 92 (1980) 24714).

 IOP Conf. Series: Earth and Environmental Science 43 (2016) 012047
 doi:10.1088/1755-1315/43/1/012047

[16] Stojanovic Z K, Jerevic S and Jovanovic 2000 Starch / Stärke 52 413.

[17] Kisung Kwon P, Joong Hyuck Auh J W, Kwan Hwa Park S, Chan H P and Cheul Jong Ko K D 1997 Starch / Stärke 69 499.

[18] Bhattacharya D, Singhal R and Kulkarni P R 1995 Carbohydr. Polym 27,167.

[19] Bochkov N P 1989 Human heredity and the environment mutagens Medicine. Vol. 8 pp. 163-167.

[20] Kozak M F 2007 Drosophila - the model of genetics: study guide (Astrakhan: Astrahan University Press) pp. 8.