

# Responses of bottom invertebrates to pollution in the Arctic: bioassays with blue mussel *Mytilus edulis* L.

A. V. Gudimov<sup>1,\*</sup> and S. S. Malavenda<sup>2</sup>

<sup>1</sup>Murmansk Marine Biological Institute of the Russian Academy of Sciences, 17, Vladimirskaia Str., Murmansk, 183010, Russian Federation

<sup>2</sup>Murmansk State Technical University, 13, Sportivnaya Str., Murmansk 183010, Russian Federation

**Abstract.** Environmental control in the Arctic ecosystems needs to be changed to hi-tech monitoring technologies with IT systems for actual and ecologically effective 'arctic safeguard'. In circumstances of the fragile arctic ecosystems, priority bioassay of possible toxic hazards in natural seawater must be undertaken. It was considered useful to examine toxic responses of some indicator species. Bivalves are well-known indicators of pollution stress arising from the activities of man, such as off-shore drilling and oil transportation, dredging or the release of pollutants. Toxic effects of some drilling fluids on energy balance, behavior and survival of the common mussel *Mytilus edulis* L, were estimated in long-duration bioassays of about 30 days each. The highest sensitivity of behavioral responses to any changes in chemical water composition was found. Mussels of the Barents Sea had significant differences from the control by four behavioral parameters even at 0.01 g/l of the ferrochrome-lignosulfonate drilling fluid, while the significant differences from the control in oxygen consumption and filtration rates were observed only at the 10 g/l. Even small concentration of another standard water-based drilling fluid with barite (of about 0.05 g/l) had a lethal effect on *M. edulis*.

## 1 Introduction

Environmental impact in the Arctic marine ecosystems is associated with increasing human activity on the shelf and seashore related mainly to exploitation of the oil and gas deposits, transportation and transshipments of mineral raw materials, including oil and condensed gas, with following harbour and terminal operations.

Standard system of environmental monitoring is outdated and failed for safeguard of the aquatic ecosystems, first of all because of its extremely delayed response to any environmental changes and pollution [1, 2]. The solely approach to reliable and advanced environmental control is online biomonitoring (OBM) technology [1, 3] with recently appeared OBM systems [1, 4]. Theoretically as a procedure, online-biomonitoring is raised up from bioassay experiments, especially performed in conditions closed to the natural ones (e.g. in semi-natural conditions).

In circumstances of the fragile arctic ecosystems, priority bioassay of possible toxic hazards in natural seawater must be undertaken.

Effects of drilling activities with accidental discharge of drilling mud in areas of oil and gas production and

possible dumping of drill cuttings in the coastal zone of the Arctic Seas are expected among others.

Drilling and dredged material often contains toxic components that are able to pollute the coastal environment and damage the highly productive area of the sea; consequently, fisheries, aquaculture, wildlife habitats and water quality are at risk.

The bioassays of toxic effects on dozens of species are not available. Hence, it was considered useful to examine toxic responses of some indicator species.

Bivalves are well-known indicators of pollution stress and can be found in aquatic habitats ranging from oceanic to freshwater. Some bivalves are often subjected to stresses arising from the activities of man, such as off-shore drilling, dredging or the release of pollutants.

Initially, the toxic effects of some drilling fluids on energy balance, behaviour and survival of the common mussel *Mytilus edulis* L, widespread along the coast of the Barents Sea, have been estimated in our long-duration tests.

## 2 Materials and methods

The main experiments were carried out at the aquaria laboratory of the Murmansk Marine Biological Institute

\* Corresponding author: [alexgud@mail.ru](mailto:alexgud@mail.ru)

located on the shore of the Barents Sea (Dalnie Zelentsy, Murmansk region, Russia). To be closer to the natural environment and real animal responses the seawater was pumped directly from the Dal'naya Zelentskaya Bay to the main laboratory tank and immediately distributed from there to aquaria.

The most part of bioassay as well as the previous background research of the coastal bivalve's activity in regard to the fluctuations of environmental factors (temperature, salinity, light, seston amount, etc.) in natural sea water flow have been explored in terms of behavioral and physiological responses in blue mussel (*Mytilus edulis*). Then some effects of environmental factors have been proved in the natural conditions of the Barents Sea and some bioassay responses to toxic substances have been verified in stationary conditions [4, 5–6].

Behavioral measurements were made by several parameters on monitoring of shell moving activity (gaping) of mussels with an automatic recorder or SG-valvometer (strain gage or tension sensor).

To measure the oxygen consumption rate with standard oxymeter, animals after the acclimation in aquaria, were placed individually or in pairs depending on their size, in glass respirometer flasks for exposures. The rate of oxygen consumption was determined from the rate of the oxygen tension declining in the respirometers sealed for the exposure time.

Filtration rate in the blue mussels was estimated by the classic indirect method [7], when the filtration rate is calculated from the decrease in particle concentration of experimental medium per unit of time. As a suspended

material, a pure culture of the unicellular green alga *Dunaliella salina* was used in our experiments. The algae were cultured according to the method described by Winter [7].

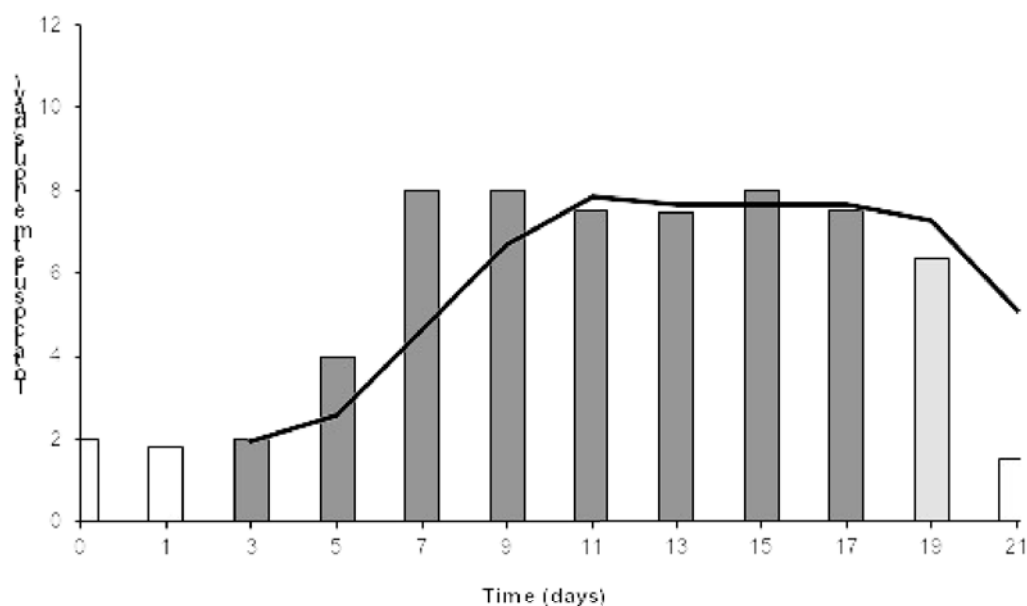
Long-term experiments presented here on bioassays of ferrochrome-lignosulfonate (FHLS) drilling fluids were performed in flow-through or stationary conditions for about a month each (28–31 days).

In addition, toxic effects of another standard drilling fluid (with barite, modified clay powder and carboxymethyl cellulose) and an experimental drilling fluid on the survival of *M. edulis* from the Kola Bay were tested in constant conditions [6]. A feature of the experimental drilling fluid (“eco-mud”) was a supplemental addition of the ground brown algae *Saccharina latissima* (past *Laminaria saccharina*) to the ferrochrome-lignosulfonate (FHLS-M) drilling fluid.

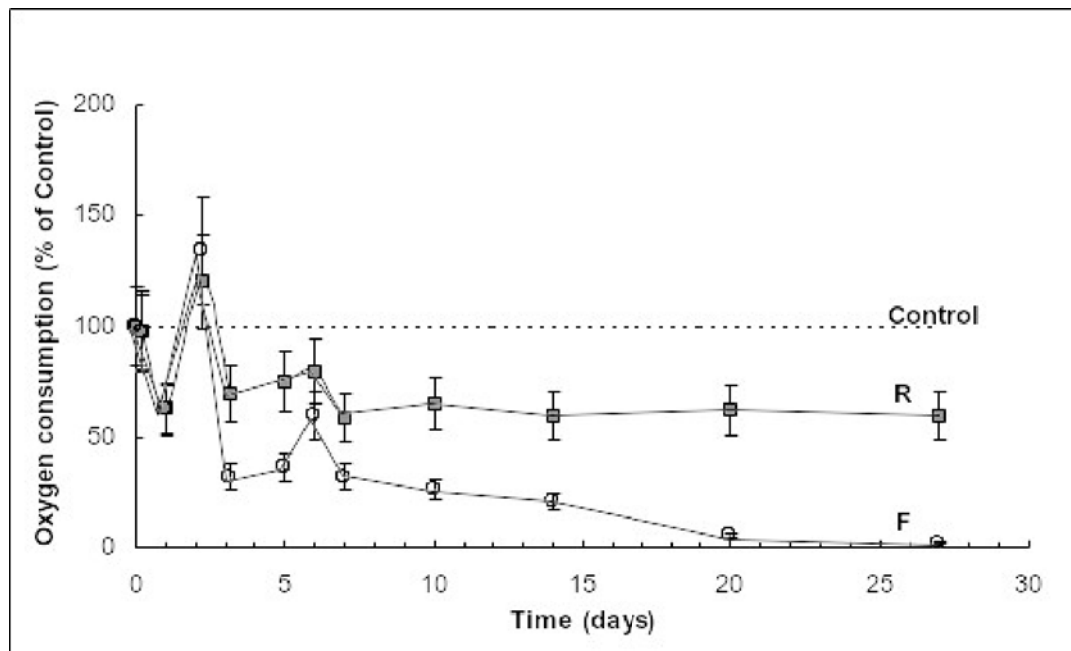
### 3 Results and discussion

The highest sensitivity of the behavioral responses in mussels to changes in chemical water composition was found in our bioassays. Mussels of the Barents Sea had significant differences ( $P < 0.05$ ) from the control by gape size-index even at 0.01 g/l of the FHLS drilling fluid (DF), while the significant ( $P < 0.05$ ) differences from the control in oxygen consumption and filtration rates were observed only at 10 g/l concentration of DF (Figures 1–2).

The time course of oxygen consumption and filtration rates in mussels exposed to DF of 10 g/l was typical for any stress response, i.e., it was followed the harsh and jog changes immediately after add of DF.



**Fig. 1.** Daily averaged total closure time in mussels during bioassay with 0.01 g/l concentration of DF (dark bars).



**Fig. 2.** Oxygen consumption (R) and filtration (F) rates in mussels under 10 g/l concentration of the ferrochrome-lignosulphonate drilling fluid (DF).

Three main phases of a typical acute response to the toxic stress at both concentrations may be distinguished here (Figure 2), including over- or undershoots of rate functions [8]. As for the mussel behaviour it has previously been reported [9, 10] that the immediate response of mussels to environmental stress is closing their shell valves followed by infrequent opening for a short time; either the animal then recovers to normal ventilation or it remains closed and dies. During this initial period of ‘shock’ when valves are closed tightly, aerobic respiration is reduced or ceases altogether [11].

At the beginning of the exposure in DF an initial decrease and then sudden and abrupt increase in the respiration and filtration rates was observed, stabilizing after a drastic decrease at a lower steady-state level of metabolic rate within 14-16 days of the exposure. The final decreases in the oxygen consumption and filtration rates were 40% and 96% of the control, respectively.

The oxygen uptake and filtration rate in *M. edulis* at 10 g/l and 5 g/l of DF strongly declined during the 28 days of the monitoring period but they were significantly ( $P < 0.05$ ) lower only at 10 g/l of DF than in the control groups. However, no significant difference ( $P > 0.05$ ) existed between the respiration and filtration rates of mussels at 5 g/l concentration of DF and in the control (0 g/l).

On the other hand, the effects of both DF concentrations resulted eventually in the death of 30 and 70% of mussels during a month of deacclimation stage (on the clean seawater flow) after the exposures in 5 and 10 g/l respectively. Hence, despite the sublethal effects obtained in the DF exposures the minimal lethal concentration of DF might be assumed as about 1-5 g/l.

In bioassay with behavioral responses of *M. edulis* the results provided clear evidence that even at 0.01 g/l

concentration of DF the behaviour of mussels was significantly affected, indicating that this concentration is high enough to be detected by the animal sensing system.

The pattern of the valve moving activity under DF exposure was abnormal and the adduction/gaping cyclic movements were performed in an abrupt, clear-cut manner. While the average gaping decreased (table 1) the total daily closure time increased significantly ( $P < 0.001$ ) in course of the experiment up to 400% of the average control level (Figure 1).

These daily periods of lasting closures amounted to at least 5 h d<sup>-1</sup>; the longest duration was obtained within a week from the beginning of the exposure to DF.

After some days of the exposure the total frequency of adductions and quiescences decreased significantly, from 26-30 to 9-11 contractions (partial closing) per day, but then increased slowly within a week of the exposure up to 15-18. This change occurred as a result of shortening the active periods and increasing the number of rest periods, resulting in the decrease of total adduction (frequency of the mussel valves closure contraction) coupled with an increase in the frequency of the alternation of active and rest periods.

The valve movement amplitude decreased twice for the first week of the exposure and then increased later up to the control level. The initial, extremely high value of the daily lasting closures slightly decreased within the exposure but down to a higher level than in the control.

There was a drastic increase of the shell gape size in mussels returned to the clean seawater flow when DF was washing out since the 18th day of the experiment (Figure 1).

**Table 1.** Average shell gape size (% of the complete shell closure) in mussels (*Mytilus edulis*) subjected to 0.01 g/l concentration of the ferrochrome lignosulphonate drilling solution (DF) for 2 weeks of the exposure: n.s., not significant at the  $P < 0.05$  level.

Conditions	X	SD	ANOVA (t- test)
Control (C)	52.4	13.44	(C - 1DF) n.s.
1 <sup>st</sup> exposure day in DF (1DF)	51.4	15.26	(1DF - 7DF) $P < 0.001$
7th exposure day (7DF)	71.6	16.20	(C - 7DF) $P < 0.001$
15th - end exposure day (15DF)	37.8	25.12	(C - 15DF) $P < 0.001$

All these changes in valve movements are integrated in the average shell gape size of mussels (table 1). Being based on series of the long-term records, we concluded that this main behavioural parameter means the degree of the bivalve organism contact with the media, and it strongly depends on the quality of the environmental conditions.

As generally considered, the toxic effects of FHLS drilling fluids are related to heavy metals of chromium and iron contained in this DF. The maximum permissible concentration (MPC) of ferrochrome lignosulfonate was estimated about 0.002 g/l [12]. The FHLS in seawater may reduce the phytoplankton production, fish embryos survival, and it impedes the development of organs and tissues in fish larvae [13]. On the other hand, at our test [6] concentrations of a modified ferrochrome lignosulfonate (FHLS-M) in the experimental drilling fluid over 5 and even 10 times of MPV did not affect the survival rate of *M. edulis*. Likely the toxic effect of FHLS-M was so mitigated because of the sorbent effect on FHLS-M by the ground algae and clay powder of this “Eco-mud”.

Toxic effects of another drilling mud tested in experiments of S. S. Malavenda [6] indicated that despite the previous estimation that barite was a low toxic substance [14], even small concentrations of the standard water-based drilling fluid with barite (of about 0.05 g/l) had lethal effects on *M. edulis*. It can be suggested that only because of the environmental or experimental conditions some aquatic organisms were not affected by barite [14], and the barite concentration of 0.5 g/l did not cause toxic effects on bivalves [15].

In some experiments with low barite concentration the filtration process and gill apparatus were damaged [12, 16]. Besides, the presence of barite enhanced the negative effect of carboxymethyl cellulose, – a component of this standard drilling fluid [12, 15, 16]. In any conditions the most pronounced toxic effect was manifested at chronic pollution [12, 16].

In our experiments carried out on blue mussels [6], the barite content in water from 0.05 g/l to 0.5 g/l, was lower or corresponded to the MPC. In spite of that all mussels died for 9 days of the exposure at any concentration of the standard drilling fluid but were alive in the control.

## 4 Conclusion

The results of our bioassays revealed that blue mussels possess a sensitive detection system and behavioral mechanism for the avoidance of pollution stress. This mechanism is responsible for both survival and permanent physiological activity of mussels under fluctuated conditions coupled with hazardous levels of contaminant in the environment

A close correlation between the duration of rest periods (valves almost or full closed) and level of contamination has been observed.

Shortening the active periods, increasing the rest time, and then alternating the active and rest periods is the main mechanism of regulation for increasing oxygen demand caused by the pollution stress.

Important to note, that the first sublethal response in blue mussels is the significant change in their shell moving activity and gaping (behavior). Behavioral changes followed by physiological responses such as the rate of oxygen consumption, filtration or cardiac activity with their significant changes at higher concentration of toxic substances.

Starting with behavioral responses the toxic stress ends up with the affected survival indicating the most harmful or lethal conditions.

Priority setting of hazards in water is the main advantage of using bioassay as a means for prevention of environmental impact and ecosystems degradation in the Arctic.

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