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2008-2022**

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**TOXICOLOGY RESEARCHES AND MONITORING OF GRAY WHALES OFF
CHUKOTKA PENINSULA (RUSSIA), 2008-2022**

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Chukotka scientists collected samples from organs and tissues of Gray whales *Eschrichtius robustus* after aboriginal whaling and landing by Chukotka Natives (Table) in 2008-2022.

Necropsy samples for heavy metals analysis included muscle, kidney, liver, blubber by 200-300 g (7.0-10.5 ounce) and blood 50-100 ml (1.7-3.4 fl.ounceUS). All samples were frozen at -24°C (-11.2F) and delivered to Laboratory of Applied Ecology of Vladivostok and to Veterinary Centers of Petropavlovsk-Kamchatsky and Anadyr (Russia), which have different measuring equipment. The preparation of tissue and organ samples for measuring performed by acidic mineralization method using nitric acid following GOST 26929-94 [1].

Chukotka Natives consume intestines and meat of whales and walruses. The RSSEHR - Russian State Sanitary, Epidemiological and Hygienic Requirements [2] are controlling and limiting the level of toxic elements of As, Cd, Hg etc. in the marine mammals' tissues and organs.

Concentrations of heavy metals and radioactivity levels never exceeded the MPL in the studied intestines samples of Gray whales in 2015-2022 (Table).

Table – Heavy metals' concentration (mg/kg mass) and radioactivity (Bk/kg) of organs and tissue samples of Gray whales, landed in the Mechigmensky Bay (Western Bering Sea), 2008-2022

		2008 ⁱ	2010 ⁱⁱ	2015 ⁱ	2016 ⁱ	2017	2019 ⁱⁱⁱ	2020 ⁱⁱⁱ	2021 ⁱⁱⁱ	2022 ⁱⁱⁱ	MPL ^{iv}
Hg	mean ± SD	0.006±0.006	0.063±0.080	0.030±0.014	0.034±0.022	0.027±0.041	0.188±0.038	0.051±0.010	0.017±0.007	0.033±0.006	0.5
	range	0.001-0.022	0.007-0.120	0.014-0.048	0.007-0.087	0.000-0.189	No data	0.029-0.060	0.005-0.030	0.004-0.058	
	N of samples	14	2	5	19	37	10	15	30	27	
As	mean ± SD	1.178±0.810	0.255±0.186	3.000±1.042	0.262±0.311	0.334±0.424	0.006±0.003	0.021±0.009	0.034±0.009	0.049±0.017	5.0
	range	0.170-2.760	0.020-0.600	1.800-4.000	0.030-1.310	0.010-1.860	No data	0.010-0.033	0.012-0.046	0.015-0.1	
	N of samples	14	13	5	19	37	10	15	30	27	
Cd	mean ± SD	0.055±0.088	0.287±0.232	0.054±0.098	0.041±0.046	0.053±0.075	0.031±0.012	0.029±0.005	0.063±0.012	0.049±0.015	0.2
	range	0.005-0.317	0.007-0.700	0.005-0.230	0.003-0.155	0.001-0.289	No data	0.017-0.036	0.034-0.083	0.010-0.080	
	N of samples	14	14	5	19	37	10	15	30	27	
Pb	mean ± SD	1.011±1.404	0.204±0.116	0.206±0.111	0.099±0.098	0.027±0.026	0.24±0.09	0.035±0.011	0.233±0.167	0.030±0.009	1.0
	range	0.070-4.200	0.053-0.450	0.100-0.390	0.011-0.344	0.000-0.160	No data	0.005-0.059	0.019-0.060	0.012-0.11	
	N of samples	14	14	5	19	37	1	15	30	27	
Radioactivity, Bk/kg mass											
Cs 137	mean ± SD						3.05	5.971±2.160			130.0
	range						No data	3.9-10.2			
	N of samples						1	7			
Sr 90	mean ± SD							4.586±1.366			100.0
	range							3.4-7.5			
	N of samples							7			

ⁱ Kovekovdova et al., 2017 [4]

ⁱⁱ Tsygankov, 2015 [3]

ⁱⁱⁱ data of Rospotrebnadzor

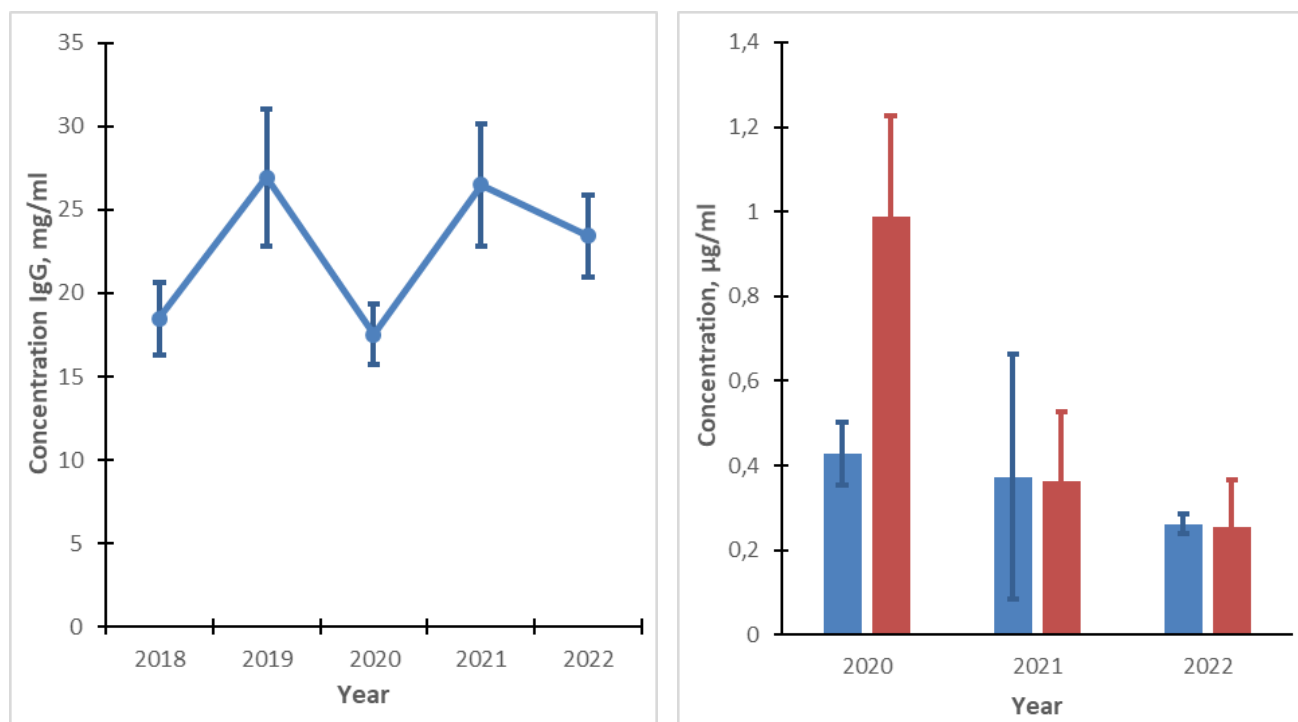
^{iv} Maximum permissible levels

Number of Gray whales in Mechigmsky Gulf was 28.3 per day in 2022 (14.3 in 2021), and a multiannual level firstly from 1999 showed a constant elevation within 38 years period of counts.

In 2022 in the Severtsov Institute the serum-positivity to nine pathogens (*Toxoplasma gondii*, *Mycoplasma* sp., *Trichinella* sp., *Candida* sp., *Chlamydia* sp., Papilloma, Herpes, Morbillivirus and Parvovirus) was determined for serum in 45 gray whales, harvested in Chukotka at 2018-2022, all of which were serum-negative to Herpes and Papilloma virus. The highest positivity level they had to *Toxoplasma gondii*, from 10% in 2020-2021 to 30% in 2022. Also *Candida* sp. level was no more than 3% in 2020-2022.

It is still unclear how negative could be the *Toxoplasma* infection for the gray whale and other marine mammals. *Toxoplasma* is an important cause of mortality in sea otters, leading to the development of encephalitis; also it causes the fatal development of toxoplasmosis in Pacific harbor seals. Isolated cases of lethal toxoplasmosis in marine mammals are often associated with animal immunosuppression as a result of morbillivirus infections. *T. gondii* infection can also kill marine mammals, affecting their behavior and increasing the risk of injury and death from predators and marine mammal hunters. Apparently, the consumption of gray whale meat by Natives can pose a certain danger due to infection with *Trichinella* and *Toxoplasma* with insufficient heat treatment of whaling products.

In addition, this year for the first time the Gray whale serum-immunity analysis was carried out, which revealed rather high rates, which varied only slightly over the years. It is quite interesting that from 13 serum-samples the concentrations of lysozyme in three "stinky" whales were minimal. It is possible that the time elapsed since the harvest of a whale may have a significant effect on lysozyme concentrations; however, this hypothesis needs to be tested. The average concentration of lysozyme in gray whales was 46.9 ± 23.9 $\mu\text{g/ml}$ serum.



Average level of lysozyme (LEFT – **blue lines**), progesterone (RIGHT – **blue columns**) and testosterone (**red columns**) in serum of Gray whales harvested in Chukotka, 2018-2022

Analysis of cortisol in the tissues of harvested Gray whales showed a multiple increase in the content of the stress hormone for animals in 2021 (more than 45 mg/ μg), which was likely due to increased killer whale activity in 2021. In 2022, increased ice conditions in the southern part of the Chukchi Sea led to a high concentration of Gray whales in the Bering Strait area, which may also influence the stress factor and cortisol concentration reached annual level of about 20 mg/ μg .

The relatively low level of sex hormones in the blood of gray whales is apparently associated mainly with the whaling on immature animals. Progesterone concentrations did not exceed 1.61 ng/ml, and testosterone concentrations did not exceed 2.74 ng/ml. No significant differences were found in

different years.

During last 5 years, wide international team worked on the “stinky” whale phenomena [5]. Since 2003, the problem of “stinky” whales arose immediately, as the meat of some harvested species possessed a strong medicinal/chemical odour. The hypotheses explaining the phenomenon ranged from biotoxins, to oil spills. To understand the problem, various tissues of normal and stinky Gray whales were collected and analyzed using headspace solid phase microextraction with Gas Chromatography – Mass Spectrometry. It was shown that dozens of smelly organic compounds were identified among over 500 compounds detected in the samples. The most interesting analytes related to the off odour are bromophenols. The most probable suspect is 2,6-dibromophenol with strong iodoformic odour, perfectly matching that of the “stinky” whales. Quantitative results demonstrated its levels were up to 500-fold higher in the “stinky” whales’ tissues. The source of 2,6-dibromophenol is likely polychaetes, producing 2,6-dibromophenol and colonizing near shore waters where whales feed [5].

Nevertheless the absence of skinny Gray whales during coastal counts and harvest monitoring, their good body conditions and stable prey content in whale stomachs, as well as other researches in period 2013-2022 indirectly indicate that the existing aboriginal Gray whales hunting has no negative impact to its population and feeding conditions in Chukotka waters, and thus can be implemented in the future in the same amount.

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