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### RESEARCH ARTICLE

#### ECOLOGY-GENETICAL EVALUATION OF RADIATION AND OIL POLLUTION (ASSOCIATED WITH HEAVY METALLS) ON BIOTA AND MAN. REVIEW.

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#### Abstract

We know that environmental factors such as pollution lead to serious genetic problems. Structural, morphological and chromosomal abnormalities were observed in terrestrial and aquatic animals of Caspian Sea region. According to research, levels of oil pollution and radiation near spills and local settlements significantly increased from 1.65 to 13.8 times nowadays. Air pollution and carcinogenic toxic substances in water, soil cause hepatitis, respiratory diseases and tuberculosis in local citizens. Patients with skin, respiratory tract, esophagus and bladder cancer are those, who live near contaminated area and contacted with oil products.

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#### Introduction:-

Ionizing radiation is a factor leading to chromosomal anomalies, as well as all types of radiation induce chromosome aberrations in the germ and somatic cells of humans [Pomerantseva M.D. et al, 1993]. Japanese scientists have studied the chromosomes in blood leukocytes of people, exposed to radiation in the atomic bombings of Hiroshima and Nagasaki [Awa and others, 1978]. Results have shown that chromosomal rearrangements of white blood cells are still present in citizens genome, even after three decades passed. Cytogenetic analysis of white blood cells of exposed people were held. According to results, all the people had irradiated leukocytes of which more than 10% had chromosomal rearrangements.

Epidemiological and experimental studies suggest induction of genomic instability in the offspring of parents exposed to ionizing radiation. Instability is primarily manifested by increased rates of mutation and cancer as well as other disorders in the offspring [Khudoley V.V., 1994]. Many research scientists have shown that the phenomenon of genomic instability found in the distant progeny of irradiated populations. It is also noted the emergence of different types of chromosomal aberrations in cells and increased the overall level of chromosomal disorders. Antypenko E.N. et al [1990] experimentally found that prolonged low-intensity radiation is able to cause a significant increase in the number of cells with chromosome aberrations and reciprocal chromosome translocations, not only in exposed, but their descendants of the first, and especially - the second generation.

Nowadays, cytogenetic study have shown the diversity of chromosomal aberrations, especially, double DNA strand breaks caused by gene-toxic agents. The population living in the region of Semipalatinsk nuclear test site have 90.0 cSv level of blood-forming organs and blood diseases, which is 2-2.5 times higher than dose at control areas (7.0-

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35,0 cSv). According to research, extending disease is iron deficiency anemia the level of which is irradiated in a dose 90-199.0 cSv1, 5-2 times higher than in the control group.

Activities done to improve the health of local populations:

-prevent anemia among women and 264,000 children under 5 years in the areas affected by nuclear test, by improving the knowledge of the pathogenesis of anemia and streamlining supply

-large-scale prevention of anemia among women and children in the region of SNT–based on rational and nutrition iron-therapy

-cytology morphometric study of peripheral blood for anemia of pregnancy to develop early diagnostic and prognostic criteria of anemia and risk allocation

-analysis of the intracellular mechanisms of anemia of mother and child

Major challenge of today is assessment of environmental impact products of nuclear explosions on the status of the human population and the development of diagnostics, health correction and rehabilitation.

**This requires:-**

- the assessment of the genetic risk of long-term effects of nuclear testing on the gene pool of human populations
- diagnosis of the physiological state of the body of people who have been influenced by years of nuclear tests.
- diagnosis of the mental state of the body of people, who have been influenced by long years of nuclear tests
- rehabilitation of health detoxification using antioxidants.

In 1986 we frequently observed aberrant in color morphs of water vole in mountain and forest area of Karkaralinsk village of Karaganda region. According to results, melanistic forms are homozygous for the recessive allele and resistant to degradation. Animals with hair dryer spot, on the contrary are less viable and fertile than animals with "normal" genotype. Cytogenetic monitoring was also performed on the population of Kazakhstan (Karaganda, Semipalatinsk, Atyrau, Aktobe regions). The purpose of monitoring is the evaluation of radiation impact and other harmful factors for hereditary apparatus. Various abnormalities in structure of the studied populations of species, including organism (morphology) and genetic (chromosomal) levels were observed in this area. In the period of 1993-2000 years the quantitative relationship and the range of structural and numerical chromosome aberrations of 35 people from the same region of Karaganda, adjacent to Semipalatinsk nuclear test site were studied. Also, 201 children, including 81 with prenatal damage of the nervous system, 70 children with birth defects, 50 healthy children (control group) were surveyed.

The results of epidemiological and experimental studies detected the induction of genomic instability in the offspring of parents exposed to ionizing radiation. This genomic instability is primarily manifested by increased rates of mutation and an increased risk of cancer and other disorders in the offspring.

Gene changes are the main category of mutations, which are known from the literature. So, 50 % of spontaneous abortion is appearance because of dominant mutations. Newborn to 96% of hereditary defect cases is caused by gene mutations. In 4% of cases, there is the action of chromosomal changes in the form of violations of chromosome number or structure [Bochkov N.P., Kuleshov N.P. et. al, 1975].

After the accident at the Chernobyl nuclear power plant a sample survey of children living in near the area was carried out. The results indicate a statistically significant increase in individual and group frequencies of aberrant cells and different types of aberrations in the children's age groups, who's living in areas with high background radiation [Shevchenko V.A. et al, 1989]. As a result of cytogenetic examination of Ukraine's population living in the area of radioactive fallout from the Chernobyl nuclear power plant were found that people significantly increased the incidence of chromosomal aberrations, particularly markers of radiation exposure (dicentrics, ring chromosomes and atypical). It is almost 8 times higher than the reference level for these mutations, accounting for 0.4 and 0.44 per 100 metaphases (control 0.05 per 100 metaphases) [Pilinskaya M.A. et al, 1993].

Pollution of the Caspian sea is the particular concern for Kazakhstan, especially its Northern and Eastern areas, where domestic and industrial wastes are discarded from Russia and Kazakhstan by rivers Volga and Ural. The

increase in oil pollution in the coastal zones of the Northern and Middle Caspian sea is to an average of 0,282 mg/l, which is the magnitude of 5-6 MPC. The maximum value of the oil pollution reaches 0.56 mg/l or exceeds the standard 11 times. Phenol concentrations reached 360 MPC (Limited concentration). Very high concentrations of petroleum hydrocarbons are contained in the bottom sediments. In shallow water (depth 10 – 15 meters) it's value amounted to 0,134-0,635 mg/kg; sediments taken from a depth of 20-100 meters of the most contaminated, contain 10963-6,570 mg/kg.

Nowadays, the state of the ecological system of the Caspian Sea and the coastal zone is characterized as extremely unfavorable. In the short term, intensive development of oil and gas fields on the continental shelf of the Caspian (marine aquatic place Kashagan) may dramatically increase the environmental burden, not only for the Kazakhstan part of the Caspian region, but throughout the area of middle and northern part of the Caspian Sea.

Oil is an important resource of polycyclic aromatic hydrocarbons (PAHs). PAHs are a broad class of stable organic compounds, which are ubiquitous in the marine and terrestrial environments. Containing more than 3 benzene rings they tend to accumulate and be retained in the benthos as a result of their low water solubility. PAHs themselves are relatively inert molecules, and it is generally assumed that the toxic and carcinogenic effects of PAHs caused by their metabolic products. Identification and enumeration of PAH and their metabolite's are objective methods for assessing environmental risk. Pyrene is always present in the test samples of oil (PAHs) and one of the most prevalent PAHs in the environment. So, pyrene may be used for assessing the carcinogenic risk of PAHs to the body, the bioavailability, and biodegradation [6, 7, 8]. Aquatic and terrestrial inhabitants were used as biological indicators. They were widespread in the study area of the Caspian Sea and Ural River. The analysis of the ability of aquatic organisms to accumulate oils and heavy metals were observed in this experiment [6,9].

### **Materials and Methods:-**

Cytogenetically methods for investigating chromosomal mutation on somatic cells of mammals by Preston et.al.,(1987) and of fish by method Vasiliyev (2009 ) were conducted in the experiment. The content of benz(a)pyrene in the body of test-objects was count. Methods of sample preparation for chromatography of polychaetes (*Nereis diversicolor*) generally developed and reproduced by us. The resulting sediment is mixed with pyrene by adding the necessary amount of pyrene dissolved in a minimum amount of acetone and mixed in the liquid portion of sediment and artificial seawater. Sediment mixed with pyrene on an automatic mixer, continuously for 5 hours. Then, after the sediment's deposition and water decant it is exposed at 5 ° C for one week prior to use in the experiment. Marine worms were kept in the sediment for five days. Worms then took out of the sediment retained in sea water to clean the bowl, at least 4 hours prior to extraction. Preparing samples of intestinal tissue, which is weighed and transferred to a test tube. In the presence of methanol, the sample is homogenized, solicited (10min) and centrifuged( 3000r / min, 5 ° C, 10min) to precipitate particles of intestinal tissue. The supernatant was filtered and transferred to vessels for high-performance liquid chromatography (HPLC), i.e. HPLC with fluorescence and UV detectors to determine the metabolites of pyrene. The results are shown in Table 3. By spectrophotometry atomic absorption method has analyzed of heavy metals content in organs and tissues of one species of fish (*Bramus brama*), sea worm (*Nereis diversicolor*), two mollusks species (*Dreissena polymorpha*, *Unio pictorum*) were done.

### **Results:-**

Objects of research: *Nereis diversicolor*, *Abramis brama*, *Unio pictorum*, *Dreissena polymorpha*. Also carried out determination of heavy metals associated with oil in marine worms (*Nereis diversicolor*), the results of which are shown in Table 1.

**Table 1:-** Content of heavy metals in marine worms-polychaetes species *Nereis diversicolor* (2009)

Sample	Linkage (g)	Cu(mg/kg)	Cd (mg/kg)	Pb(mg/kg)	Zn(mg/kg)	Fe(mg/kg)	Ni(mg/kg)	Sr(mg/kg)
( <i>Nereis diversicolor</i> ) fixed	0,87	6,8	0,098	92,18	30,02	350,71	3,15	7,02

**Table 2:-** Content of heavy metals in marine worms-polychaetes of species *Nereis diversicolor* (2009), 2010)

Name of sample	Linkage (g)	Cu(mg/kg)	Cd (mg/kg)	Pb(mg/kg)	Zn(mg/kg)	Fe(mg/kg)	Ni(mg/kg)	Sr(mg/kg)
( <i>Nereis diversicolor</i> ) fixed	0,87	7,3	0,91	1,3	32,2	360,3	3,8	8,01

As follows from the table the greatest ability to accumulate in the body of polychaetes is shown by iron, lead, zinc and copper. Our research has shown that the evaluation of marine worms (*Nereis diversicolor*) as a test object is the most sensitive and reliable method for the analysis of high-performance liquid chromatography with fluorescence and UV detectors.

**Table 3:-**The content of benzo (a) pyrene in the tissues of polychaetes from the northern coastal area of the Caspian Sea (2009)

N <sub>o</sub>	Object	Content of benzo(a)pyrene, mcg/kg	X <sub>m</sub> , mcg/kg
1	<i>Nereis diversicolor</i>	1,20	1,084
2	<i>Nereis diversicolor</i>	1,01	
3	<i>Nereis diversicolor</i>	1,04	

**Table 4:-**The content of benzo(a)pyrene in the tissues of mollusks from the coastal zone of the northern Caspian (2010)

N <sub>o</sub>	Object	Content of benzo(a)pyrene, mcg/kg	X <sub>m</sub> , mcg/kg
1	<i>Unio pictorum</i>	1,03	1,084
2	<i>Unio longirostris</i>	0,09	
3	<i>Unio pictorum</i>	0,02	

Investigations on the determination of pyrene and its derivatives were carried out by HPLC method. Obtained results have shown that the accumulation of benzo(a)pyrene in the body of polychaetes significantly increased by years, which indicates the intensity of oil pollution in the study area. Accumulation of pyrene components (benzo[a]pyrene) in the tissues of marine organisms allows us to predict the inclusion of PAH in the metabolism of worms. Identifying, defining and counting the number of PAHs and their metabolites is a rather objective method for the testing carcinogenicity of oil and oil-derived waxes as environmental pollutants.

**Table 5:-**The content of heavy metals in organs and tissues of two fish species and shellfish.

Samples title	Samples in gr.	Cu in mg/kg	Cd in mg/kg	Pb in mg/kg	Zn in mg/kg	Fe in mg/kg	Ni in mg/kg	Sr in mg/kg
Liver ( <i>Bramus brama</i> ) in formalin	0,43	2,57	0	66,36	8,9	149,8	0,55	5,45
Muscles ( <i>Bramus brama</i> ) in formalin	0,81	3,0	0	18,615	3,93	34,44	1,4	8,34
Gills ( <i>Bramus brama</i> ) in formalin	1,105	0,5	0	58,47	10,99	174,15	1,14	6,216
Gonads ( <i>Bramus brama</i> )	0,54	0	0	33,44	5,59	76,19	1,47	4,64

Dreisena polymorpha (soft tissues)	0,16	0,0288	0,0045	0,504	4,096	0,8	0,08	1,0
Dreisena polymorpha (sink)	0,6	1,188	0,26	9,72	1,56	40,0	4,56	1,0
Unio pictorum (soft tissues)	0,95	0,96	0,46	16,18	14,49	226,67	1,91	1,67
Unio pictorum (sink)	0,79	3,408	0,71	18,46	2,15	22,57	11,97	2,33

### Discussion:-

The progressive impact of anthropogenic factors on natural populations of animals and plants requires a detailed environmental-genetic analysis. In assessing the state of natural ecosystems, small mammals play an important role. We, therefore, conducted this study to assess the mutagenic potential danger of pollution by oil, oil products, and heavy metals, using as a test object, natural populations of the great gerbil (*R. opimus*). In particular, the studies conducted to assess the genetic risk of petroleum, petroleum products, and heavy metals in trophic food chains in natural communities of rodents in the "oil-soil-plant-animal system." As noted by Anderson (1985), the power supply is used for analyzing the structure and functioning of ecosystems.

In all investigated Zhyloysk area plants *S.nitfraria*, *T.sibirica* have the highest cumulative and sustained ability to heavy metals. These species may be used as bioindicators of environmental pollution with heavy metals. The high content of metals in the oil-contaminated areas of Atyrau region may be the result of saturation of the lower horizons of the soil or associated with ore-bearing deposit of oil and gas sectors.

According to the literature, in the oil-contaminated plant's ecotopes in natural populations lead level exceeds the background values of 2.5 to 5 times, and in our studies in the oil fields and in the vicinity of the road level lead concentrations exceed the range of 1.65 to 13, 8 times.

A.B.Bigaliev noted earlier (1986), screening methods for the study of biological systems of rodents to determine mutagenic substances for the induction of genetic alterations in the cells of small mammals *in vitro* and *in vivo*. The frequency of cells with chromosomal aberrations in the bone marrow of *R.optimus* rodents is an important characteristic of gene-toxic properties of pollution by oil, oil products and heavy metals and the intensity of mutation. A number of the cytogenetic studies of natural populations of animals were conducted. Such an approach to characterize the state of natural populations involves obtaining cytogenetic data for different types of animals to generate information for the spontaneous level violations and assess response to all kinds of negative impacts. Therefore, the results of the research in Tengiz and Kulsaryoil field and products, using *R.opimus* as a test object was showed mutagenic changes and all types of chromosome abnormalities. The maximum values of the studied cytogenetic parameters were observed in highly contaminated areas. Chromosome aberrations in the bone marrow of the test objects exceeded the reference value of 3.9 times. The correlation coefficient between the content of oil and heavy metals in soil, plants, and the frequency of chromosomal aberrations in bone marrow cells *R.opimus* is  $r = 0,97$ , between the content of oil and heavy metals in soil and plants with a level of chromatide type aberrations  $r = 0,81$ , between the oil and the level of chromosomal aberrations such as  $r = 0,96$ . The quantitative and qualitative composition of cytogenetic damage at the presence of oil products and sulfur content depends on the distance from the source of pollution in the studied areas.

The level of genetic damage to wild rodents can be considered as the real maximum mutagenic effect on environmental factors in relation to the people living in these areas. In contaminated areas as Zhylyoi district high degree of air pollution and soil carcinogenic and toxic substances causing severe population hepatitis, respiratory diseases and tuberculosis were defined. Deaths from these diseases in 1987 was in the region of 36-37% including children - 15-17%. Among patients with cancer of the skin, respiratory tract, esophagus and bladder are those who had contacted with oil and oil products. The data for 1973-1987 years shows that the incidence of malignant tumors in the Emba (current Zhylyoy) area -138.9% to 100 thousand people, in Makat-138.4% and Atyrau region was -195.3%.

**Conclusion:-**

Thus, it can be concluded that the studied mutagenic agents, heavy metals and oil are not the only reason for the high frequency of chromosomal mutations of *R. opimus* surveyed areas. It is difficult to say which of chemicals caused the observed changes. The data showed that oil and petroleum products are highly toxic and complex substances, which influence heredity of living organisms. This is reflected in the ecosystems of Zhylyoi district of Atyrau region, as well as the entire Northern Caspian region, by disappearance of individual genotypes from existing species of plants and animals. Each year the transgression and regression of the Caspian Sea affect the west coastal landscape. Therefore, at present there is a problem of sustainable biodiversity conservation, genetic resources and ecosystems are not only local, but also regional. Studies are needed not only to assess the condition of natural systems under anthropogenic stress but also to predict the mutagenic and carcinogenic risks of environmental factors for the people living in these areas. Thus, the environmental problems of oil and gas complex in Western Kazakhstan, is a serious threat to the Caspian Sea region, the scale and nature of the disaster may cause an unprecedented scale, the damage from which is almost impossible to assess.

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