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EQUI-DOSIMETRY OF DELETERIOUS FACTORS AT THE LEVEL OF POPULATIONS AND COMMUNITIES OF AQUATIC ORGANISMS

A principally new ecological tool is proposed for comparing equivalent effects of very different factors at the levels of populations and communities as the new approach - equi-dosimetry of effects by radiations (ionizing and ultra-violet), acidification and metals as well as any other deleterious impacts.

Key words: radiochemoecology, equi-dosimetry, nuclear and non-nuclear pollutions/factors, microcosm, *Daphnia*, aquatic populations and communities, environmental critical zones, major interphasic marine ecosystems, Black Sea

The present paper is a synthesis and a review of three fields of logically mutually connected studies: (I) the radiochemoecological conceptual model of effects in all possible scales of deleterious factors and the equi-dosimetry approach for ecotoxicity ranking of various toxic chemicals on the basis of ionizing radiation, which were originated and are being developed at IBSS, Sevastopol, Ukraine [15 – 19]; (II) experimental studies of ecological effects of various toxic agents on the aquatic microcosm in comparison with ionizing radiation, originally proposed and are being elaborated at NIRS, Chiba, Japan [3 – 8] and (III) long-term studies in the Black Sea ecosystems – critically impacted zones – which are carrying out on the basis of the concept of the main marine interphasic ('contouric') communities and their interactions at IBSS Odessa Filial branch, Odessa, Ukraine [23 – 25].

I. RADIOCHEMOECOLOGICAL CONCEPTUAL MODEL

One of us [14, 15] had proposed and developed a conceptual model of zones of responses of organisms, populations and ecosystems to all possible dose rates of ionizing radiation in the environment (Fig. 1). These zones are: the zone of well-being (natural background levels of environmental ionizing radiation below 0.005 Gy/y), the zone of physiological masking (0.005 – 0.1 Gy/y), the zone of ecological masking (0.1 – 4 Gy/y) and the zone of damage to ecosystems (>> 4 Gy/y).

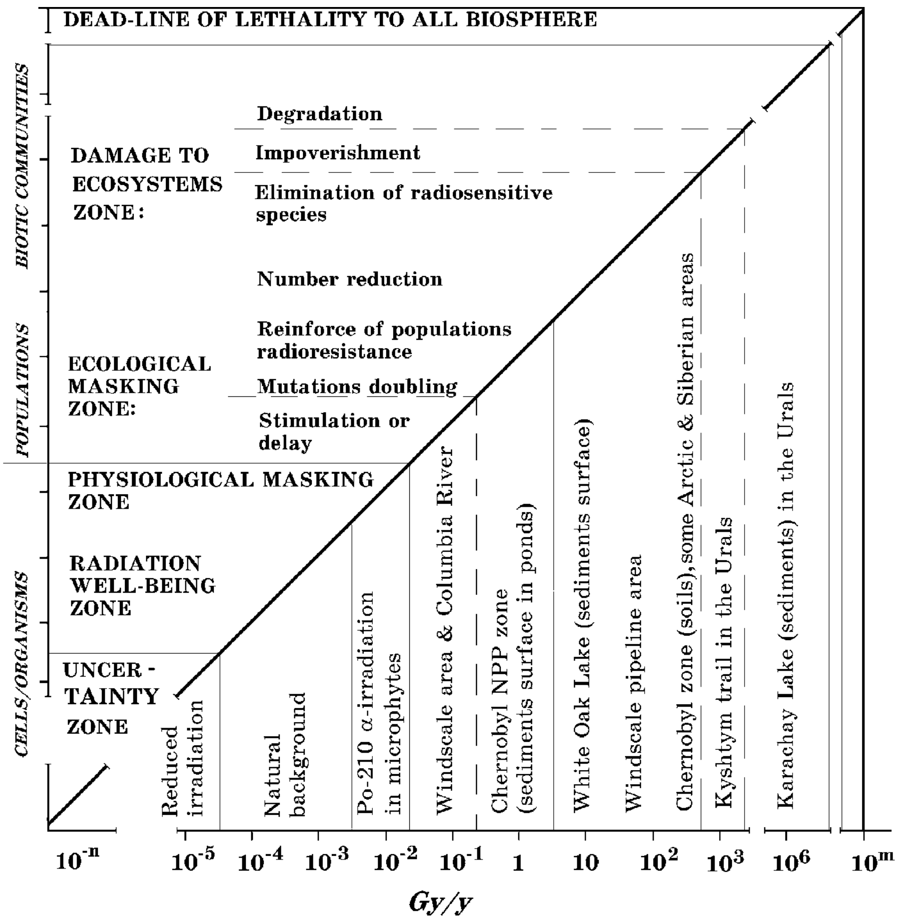


Fig. 1. Zones of dose rates and their effects in the biosphere [15, 16]

Рис. 1. Зоны мощностей доз и производимые ими эффекты в биосфере [15, 16]

The model was afterwards extended and transformed into the radiochemoecological conceptual model [16 – 19], which covers effects of both ionizing radiation and chemical pollutants (Fig. 2). The following ecological criteria are used in the Fig. 2: decrease of number or total mass (of populations), decrease of number of species (of communities) and degradation of communities (of ecosystems).

A good agreement of the microcosm data on chemical and ionizing radiation impacts [4] with the radiochemoecological model mentioned above was demonstrated [17]. Equi-dosimetry means the equal ability of proper amounts/doses of different factors (for example, ionizing radiation, UV, acidity, heavy metals a.s.o.) to produce the same quantitative effect.

Ecological Gray-equivalent of the effects of chemicals is the ratio: Gy/C, where C is a chemical concentration [19]. In cases of heavy ionizing particles we have to use Ecological Sievert-equivalent.

II. EXPERIMENTAL STUDIES

Usefulness of the radiochemoecological conceptual model and equi-dosimetric approach was demonstrated by the following examples, in which effects of various toxic agents on water fleas or the aquatic microcosm were compared with those of ionizing radiation.

II.1. Water-flea ecotoxicity test

The water flea *Daphnia* is a typical freshwater zooplankton. This organism is sensitive to toxic chemicals, and is a key group in aquatic ecosystems. That is, *Daphnia* is strong in competition among herbivorous zooplankton species,

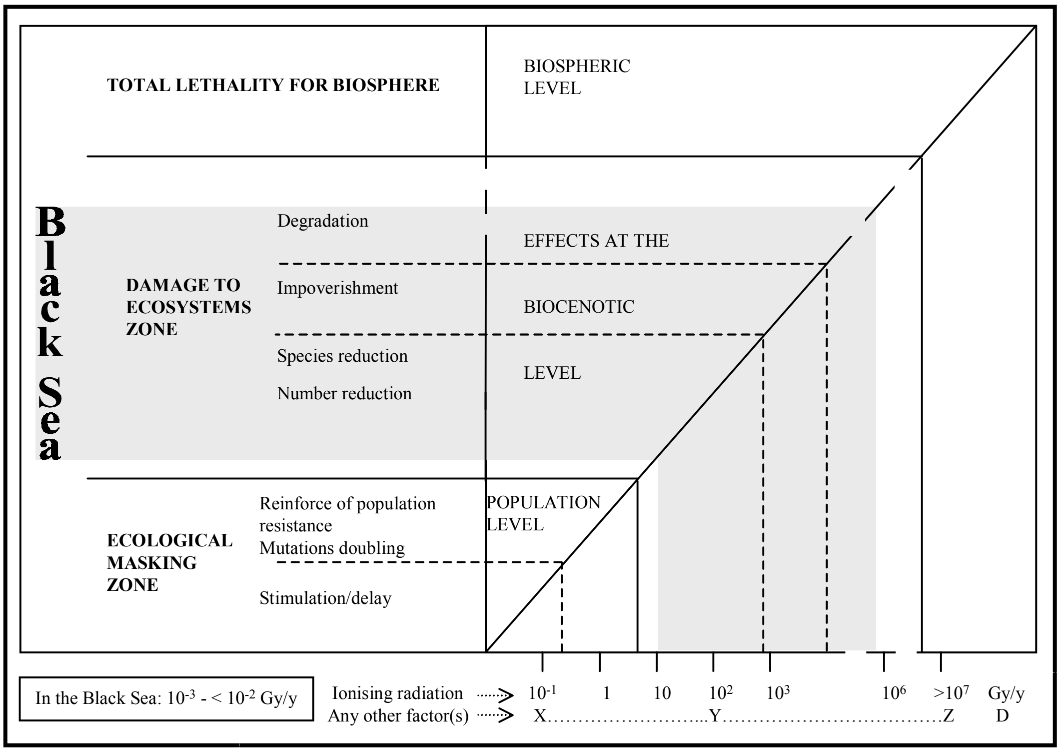


Fig. 2. Long-term reactions of populations/ecosystems to ionizing radiation as well as any chemical or physical factors at the doses in 'masking' and 'damage' zones. X, Y, Z - doses D of non-radioactive factors in each zone. Ecological impacts in the Black Sea are connected with non-nuclear factors because of low doses of ionizing radiation [17].

Рис. 2. Долговременные реакции популяций/экосистем на воздействие ионизирующих излучений и любых химических или физических факторов при дозах, соответствующих зонам "маскировки" и "повреждения". X, Y, Z - дозы D нерадиоактивных факторов в каждой зоне. Экологические последствия в Черном море связаны с неядерными факторами по причине низких доз ионизирующей радиации [17].

and appears to play a key role to control algal biomass. Therefore, once the *Daphnia* population suffers severely by chemicals such as insecticides, some other zooplankton species such as rotifers or algae are indirectly increased. For these reasons, *Daphnia* is generally used for ecotoxicity screening of chemicals, and the standard methods for this screening were proposed by the Organization for Economic Co-operation and Development (OECD) and other international or national organization [9]. We investigated effects of acute γ-irradiation and some heavy metals (manganese, nickel and copper) on the mobility of *Daphnia magna* according to the OECD method (OECD Guideline for Testing of Chemicals 202; *Daphnia* sp., acute immobilization test). The details of this study were reported elsewhere [7].

No significant effects on mobility of *D. magna* were observed at 24 hours after acute 1200 Gy irradiation, but 1350 Gy irradiation significantly immobilized this organism. The immobility rate of *D. magna* became higher with the increase in absorbed doses, and almost all individuals were immobilized at 2000 Gy. The dose-response curve was a sigmoid-shape, which is typically observed in lethality for irradiated animals such as rats, monkeys and so on. From this dose-response relationship, the median effect dose (ED_{50}), at which 50 % individuals of *D. magna* were immobilized, was estimated to be 1600 Gy (Table 1).

Table 1. Median effect doses (ED_{50}) and Gy-equivalent factors ($GyEF_D$) in the *Daphnia* immobilization test
Табл. 1. Средние эффективные дозы (ED_{50}) и Гр-эквивалентные факторы ($GyEF_D$) при тесте обездвижения *Daphnia*

	γ-rays	Heavy metals		
		Mn	Ni	Cu
ED_{50}[1]	1600 Gy	990 μM	180 μM	3.3 μM
$GyEF_D$[2]	•	1.6	8.9	480

[1] Dose at which 50 % individuals of *D. magna* were immobilized;
[2] ED_{50} of γ-rays /ED_{50} of heavy metals.

Fractions of immobile *D. magna* were positively correlated with log-transformed doses of each metal, and the relationships between them could be fitted by sigmoid curves, which are typically observed in dose-responses of organisms exposed to toxic chemicals. These dose-response relationships provided the estimation of the ED_{50}s shown in Table 1.

The equi-dosimetric approach was adopted for toxicity ranking of the heavy metals to *D. magna* on the basis of γ-rays. That is, the Gy-equivalent factors for *D. magna* ($GyEF_D$) were defined by the following equation:

$$GyEF_D = \frac{ED_{50} \text{ of } \gamma-\text{rays}}{ED_{50} \text{ of heavy metals}} \qquad (1)$$

The $GyEF_D$s of the heavy metals concerned are shown in Table 1. The lager the $GyEF_D$ value is, the higher the toxicity is. Toxicity of the heavy metals to *D. magna* could be therefore ranked as Cu>Ni>Mn.

II.2. Microcosm ecotoxicity test

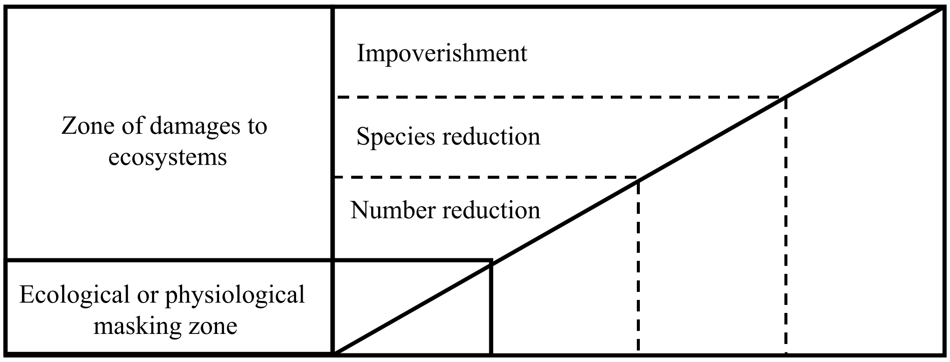
Microcosms are small scales of experimental model ecosystems constructed in the laboratory. They provide the biotic or abiotic simplicity, controllability and replicability, which cannot be expected in field studies. They also contain interspecies interactions as do natural ecosystems. This means that microcosm tests can evaluate not only direct effects of toxic agents, but also community-level effects due to interactions among the constituting species or between organisms and toxic agents, which cannot be evaluated by conventional single-species tests such as the *Daphnia* immobilization test described above [1].

We investigated ecological effects of γ-rays [3] and various other toxic agents such as ultraviolet-C radiation (UV-C) [22], acidification [12], aluminium [8], manganese [5], nickel [4], copper [8], gadolinium (Gd; [6]) and dysprosium (Dy; Fuma et al., manuscript in submission) using a microcosm consisting of flagellate algae *Euglena gracilis* as a producer, ciliate protozoa *Tetrahymena thermophila* as a consumer and bacteria *Escherichia coli* as a decomposer [10]. This microcosm mimics essential processes in aquatic microbial communities [11].

That is, the microcosm is maintained with photoenergy which *Eu. gracilis* fixes by photosynthesis. Metabolites and breakdown products of one species contribute to growth of the other two species. *T. thermophila* grazes *E. coli* as staple food. As a result of these interspecies interactions, these three species can co-exist for more than one year without addition of any nutrients. The microcosm can be therefore regarded as a self-sustaining system. The microcosm can evaluate not only direct effects of toxic agents but also community-level effects, though it is very simple. For example, 100 μM copper extinguished *E. coli* first and then *T. thermophila* in the microcosm. It is thought that this extinction of *T. thermophila* was not a direct effect of copper but indirectly arose from extinction of *E. coli* [8].

In general, degrees of effects observed in the microcosm exposed to each toxic agent were positively correlative with its dose. For example, acute irradiation by 50 or 100 Gy γ-rays temporarily decreased the cell density of *E. coli*. At 500 or 1000 Gy, *E. coli* died out, and the cell densities of the other two species were decreased compared with controls. At 5000 Gy, all species died out [3]. For another example, at 1 or 10 μM nickel, the cell densities of all species were not affected.

At 100 µM, *T. thermophila* and *E. coli* died out. At 1000 µM, all species died out [4]. Though effects observed in the microcosm were different in details among the toxic agents, the microcosm generally showed the following dose-response pattern corresponding to the radiochemoecological conceptual model: (1) "Ecological or physiological masking zone", i.e., no effects on the cell densities, but there might be some effects that did not lead to decrease in the cell densities; (2) "Number reduction", i.e., decrease in the cell densities of at least one species; (3) "Species reduction", i.e., extinction of one or two species; and (4) "Impoverishment", i.e., extinction of all species. According to this categorization, effect doses of the various toxic agents compared with ionizing radiation could be summarised as Fig. 3.



γ-rays (Gy)	ŕ |	50-100	500-1000	5000
UV-C (J/m²)	100	1000	5000	10000
Acidification	ŕ |	pH 4	pH 3.5	ŕ |
Al (мM)	10	100-500	ŕ |	1000
Mn (мM)	ŕ |	100-1000	10000	ŕ |
Ni (мM)	10	ŕ |	100	1000
Cu (мM)	10	ŕ |	100	ŕ |
Gd (мM)	50	100	300	1000
Dy (мM)	50-100	180	300-560	1000

ŕ :|Not examined

Fig. 3. Effect doses of γ-rays and other toxic agents for the microcosm evaluated by the radiochemoecological conceptual model
Рис. 3. Эффективные дозы гамма-лучей и других токсических агентов для микрокосма, определенные с помощью радиохемоэкологической модели

When "species reduction" is adopted as an endpoint, the Gy-equivalent factors for the microcosm ($GyEF_M$) in the equi-dosimetric approach can be defined by the following equation:

$$GyEF = \frac{\text{"Species reduction" dose of } \gamma-\text{rays}}{\text{"Species reduction" dose of other toxic agents}} \quad (2)$$

Table 2 shows the $GyEF_M$s of the heavy metals concerned. Toxicity of these heavy metals to the microcosm could be therefore ranked as Cu=Ni>Gd≥Dy>Mn. This toxicity rank was partly the same as that obtained from the *Daphnia* immobilization test described above. That is, both results of the microcosm and *Daphnia* tests indicated that copper and nickel were more toxic than manganese. However, there was a noteworthy difference in the ecotoxicity rank between these toxicity tests. This difference was that copper and nickel had the same toxicity to the microcosm, while copper was more toxic to *Daphnia* than nickel. It is thought that this difference in the toxicity rank resulted from different sensitivities to these metals between *Daphnia* and the species constituting the microcosm.

Table 2. Gy-equivalent factors for the microcosm ($GyEF_M$)

Табл. 2. Гр-эквивалентные факторы для микрокосма (GyEFм)

	Mn	Ni	Cu	Gd	Dy
$GyEF_M$[1]	0.05-0.1	5-10	5-10	1.7-3.3	0.89-3.3

[1] "Species reduction" dose of γ-rays /"Species reduction" dose of heavy metals

Main conclusions from experimental studies presented above are as follows. The equi-dosimetric approach makes it possible to rank ecotoxicity of various toxic agents on the basis of ionizing radiation, whose dose estimation and biological effects have been investigated more in detail than the other toxic agents. In the conventional single-species ecotoxicity test, i.e., the *Daphnia* immobilization test, the equi-dosimetric approach was useful for toxicity ranking of some heavy metals. In the microcosm test, the radiochemoecological conceptual model was useful to categorise effects of ionizing radiation and some other toxic agents.

In addition to this model, the equi-dosimetric approach made it possible to rank toxicity of some heavy metals to the microcosm at the community-level. It is therefore thought that combination of the radiochemoecological conceptual model and equi-dosimetric approach is useful for ecotoxicity ranking of various agents on the basis of ionizing radiation in complex microcosms and natural ecosystems. Such ecotoxicity ranking will contribute to a better choice of human activities for conservation of ecosystems. That is, it will provide some scientific basis for replacement of harmful activities with less harmful ones.

III. LONG-TERM MONITORING OF THE BLACK SEA ECOSYSTEMS

All available data on the fate of species, populations and communities of the Black Sea, especially in its NW shelf area, were summarized [23 – 27].

The famous great benthic community of "the Zernov' Phyllophora field" consists of four species of red (agar-bearing) macroalgae – *Phyllophora* genus. They are key species of this very rich biocenosis in the Black Sea degraded to the tragic extent under severe anthropogenic impact in 1950 – 1990. Though this degradation was created under general influence of various anthropogenic activities, the main cause was the decline of water transparency due to eutrophication and increased number of phytoplankton and detritus in water column [23 – 25] (Table 3).

Table 3. Decline of habitat areas and biomass of *Phyllophora* species

Табл. 3. Уменьшение ареалов и биомассы видов *Phyllophora*

Period, years	Inhabited area, km²	Total biomass, t
1950s	11000	10000000
1960s	7000	4000000
1980s	3000	1400000
1990	500	300000

One of dominated negative ecological factors in the large area in front of the Danube river delta is eutrophication, which causes the lack of oxygen in near-bottom layer of water – hypoxic conditions ('hypoxia').

Here is a series of data on increase in its damaged area on the NW shelf at depths from 10 to 40 m in 1973 – 1990 [23 – 25]:

Year	1973	1974	1975	1976	1977	1978	1979	1980	1981
Damaged area, 10^3 km^2	4	12	10	3	11	30	15	30	17
Year	1982	1983	1984	1985	1986	1987	1988	1989	1990
Damaged area, 10^3 km^2	12	35	10	5	8	9	12	20	40

From 100 to 200 t of benthic invertebrates and fishes per 1 km^2 died in a hypoxia period. During studied 18 years (1973 – 1990) 60 MT of bottom hydrobionts, including up to 5 MT of fishes, perished from hypoxia. Ecological situation in the NW shelf was somewhat improved in the second half of 1990s, but exact data on hypoxia areas are absent.

Severe impacts shown in Table 4 have been observed in the Black Sea shelf and estuarine ecosystems, populations and species. These impacts are quantitatively equivalent to the ecological effects of nuclear pollution/contamination in the Chernobyl NPP' nearest zone, the Kyshtym trail, the Karachay Lake, the other similar nuclear areas in the World. But dose rates of ionizing radiation to biota are less than 0.007 Gy/y in the Black Sea (Table 4). It is therefore thought non-nuclear pollution has caused the severe impacts in the Black Sea.

Table 4. Damage to ecosystems and populations of the Black Sea species caused by nuclear and non-nuclear pollutions [17]

Табл. 4. Поражение экосистем и популяций черноморских видов, вызванное ядерными и неядерными загрязнениями [17]

Ecological damage assessments [26]	Lethal Chronic Doses of Ionising Radiation, Gy [28]	Equivalent Lethal Chronic Doses of Non-Nuclear Pollutions, Gy-eq./y [A]	Environmental Nuclear Sources, Gy/y [B] [13, 20, 21]	Ratio A/B
1	2	3	4	5
1. *Cystoseira barbata* biocenosis > 99 % of its population size were lost since 1960s on Romanian & Ukrainian shelf under eutrophication & pollution impact.	For *Thallophyta* (including algae) 180 – 72000	180 – 72000	< 0.005	$10^4 – 10^7$
2. Gen. *Phyllophora* biocenosis 97 % of population size were lost during 30 years on NW Black Sea under press of eutrophication, pollution & turbidity.	For *Thallophyta* (including algae) 180 – 72000	180 – 72000	< 0.005	$10^4 – 10^7$
3. *Ostrea edulis* > 95 % of its population were lost during 30 years on the Black Sea shelf under turbidity action.	For *Mollusca* 600 – 6000	600 – 6000	< 0.007	$10^5 – 10^6$
4. *Mytilus galloprovincialis* 60 % of its population were lost during 30 years on the Black Sea shelf under action of hypoxia, caused by eutrophication.	For *Mollusca* 600 – 6000	600 – 6000	< 0.007	$10^5 – 10^6$
6. *Gobiidae* populations All 20 species were lost by 80 % of their populations on the shelf during 30 years under action of hypoxia on the bottom & destruction of their breeding places.	For *Pisces* 42 – 360	42 – 360	< 0.007	$10^4 – 10^5$

Table 3. Contnd.

1	2	3	4	5
7. Marine *Mammalia* populations Dolphins (3 species, endemic subspecies) lost by 90-95 % of their populations sizes during 30 years as consequences of toxicants bioaccumulation & killing as bycatch in fisheries. Number of monk seal (*Monachus monachus*) decreased to few individuals (or to 0) because of lack of reproduction on coastal areas & toxicants bioaccumulation.	For *Mammalia* 12 – 90	12 – 90	< 0.006	$10^3 – 10^4$

IV. SHORT SUMMARY

Summarizing all materials presented and discussed in this article, including radio-chemoecological conceptual model, principles of eco-dosimetry, results of experimental laboratory studies and long-term ecological monitoring in the sea we would like to underline that the new approach – equi-dosimetry of radiations (ionizing and ultra-violet), acidification and metals (Al, Cu, Dy, Gd, Mn, Ni) formed a principally new ecological research tool for comparing very different deleterious factors at the level of populations and communities.

At the same time we consider our work as an attempt to start to fill some 'Gaps in scientific knowledge and development requiring consideration' outlined in 'Statement of International Union of Radioecology' concerning radiation and general protection of the environment [2].

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Еквідозіметрія ушкоджуючих факторів на рівне популяцій та угруповань водних організмів. Г. Г. Полікарпов, Ю. П. Зайцев, Ш. Фума. Запропоновано новий екологічний інструмент для порівняння рівних ефектів різних факторів на рівні популяцій та угруповань у якості нового підходу - еквідозіметрії ефектів випромінювань (іонізуючих і ультра-фіолетових), ацідіфікації і металів, а також будь-яких інших ушкоджуючих впливів.

Ключови слова: радіохемоекологія, еквідозіметрія, ядерні і неядерні забруднення/фактори, мікрокосм, *Daphnia*, водні популяції та угруповання, екологічні критичні зони, важливіші контурні морські екосистеми, Чорно море

Эквидозиметрия повреждающих факторов на уровне популяций и сообществ водных организмов. Г. Г. Поликарпов, Ю. П. Зайцев, Ш. Фума. Предложен новый экологический инструмент для сравнения равных эффектов различных факторов на уровне популяций и сообществ в качестве нового подхода - эквидозиметрии эффектов излучений (ионизирующих и ультрафиолетовых), ацидификации и металлов, а также любых других повреждающих воздействий.

Ключевые слова: радиохемоэкология, эквидозиметрия, ядерные и неядерные загрязнения/факторы, микрокосм, *Daphnia*, водные популяции и сообщества, экологические критические зоны, важнейшие контурные морские экосистемы, Черное море