## **ARE THE "LOW DOSES" TRUE LOW?**

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 ABSTRACT: *The article states that the Chernobyl accident caused only a quantitative change in the pre-existing spectrum of radiobiological problems in the direction of increasing attention to the theoretical and practical issues of the effect of chronic ionizing radiation on biological objects of different levels of structural and functional organization. It is believed that chronic radiation influences biological objects in the so-called "low doses", the quantitative measure of which, however, is determined by subjectively chosen criteria. The article considers two main aspects of the problem of "low doses": firstly, to establish a relationship between these doses and subthreshold doses and, secondly, to show the role of heterogeneous tropism of biological structures in relation to their ability to absorb radionuclides from the environment, which contributes to the formation of uneven dose loads.*

 **Key words:** "low doses", "subthreshold doses", radiocesium, radiostroncium

The relationship between the concept of "low doses" and the concept of "subthreshold doses". The resulting radioecological situation determined such characteristics of dose loads on organisms that forced radiobiologists to move from studying the predominantly effect of acute exposure at doses that cause radiation sickness or deterministic effects in the general case, to a closer study of the so-called "low doses" of radiation which are associated, as a rule, with chronic exposure conditions.

Probably the most practical definition of "small doses" would be to define them as poorly studied doses due to many methodical and methodological difficulties (the complexity of dosimetry, statistical data processing, the need for large amounts of research, the complexity of interpreting the results). If we try to give a more exact definition of "small doses", it is necessary, first of all, to determine or select a test reaction by which one can judge the effect of radiation (chromosomal aberration frequency, mitotic index, average life expectancy (ALS) etc.). Obviously, most reactions will have a threshold of sensitivity and (or) resistance to radiation. In this case the only non-threshold reaction will be the ionization or excitation of atoms and molecules of the irradiated object, since the energy of quanta or particles of ionizing radiation is much greater than the ionization energy of atoms or the energy of their covalent bonds in molecules. In this regard, the question of the threshold effect of radiation at a given level of integration is removed, and in relation to these reactions it is incorrect to talk about small doses or rather it is meaningless regarding the large values of the energy of particles and quanta.

Any absorbed dose is capable of causing a response at the molecular level, and the whole problem comes down to its detection. The existence of quasi-thresholds and, accordingly, quasithreshold doses found in the study of dose dependences of the survival of unicellular organisms testifies just in favor of the fact that these reactions are non-threshold and, therefore, are caused by damage of N-impact targets, the probability of which always exists according to the Poisson nature of the distribution primary acts of interaction of quanta or corpuscles of radiation with the target. Obviously, the degree of manifestation of the reaction (reproductive death, for example) to irradiation in each cell does not depend on the absorbed dose; therefore, such reactions are usually called stochastic. This class of reactions should also include somatic (carcinogenic, for example) and hereditary effects that arise on the basis of one damaged cell but retained the ability to reproduce cells [1].

All reactions or stages of reaction to irradiation develop within the framework of a specific structural and functional hierarchy of the biological system under study (from cells to ecosystems). If they (reactions) have a pronounced threshold character, then the dose dependence has a true shoulder [2]. Only for reactions of this kind it is expedient to speak about the existence of small doses, which are more correctly (stricter) called subthreshold. Of course, a subthreshold dose for one type of effect may simultaneously be above the threshold for another effect and vice versa. For example, subthreshold doses for insects regarding the effect of irradiation on life expectancy turn out to be sterilizing or at least disrupting gametogenesis. Probably, in this case insects "survive" due to the functioning of somatic cells that are more radioresistant to radiation. In another situation a subthreshold dose in relation to the inhibition of a function may be suprathreshold in relation to the stimulation of the same or another function. In other words, we should talk about the complex potential structuring of any studied threshold.

Studying the reactions of biological objects of any structural and functional level, one can always distinguish a range of doses called "small" which are essentially subthreshold. For instance, even subthreshold doses that are close to those that induce interphase death of plant cells and are measured in thousands of *Gy* have the right to be called small, since they are not capable of inducing a transient process in a cell ending in its metabolic death. Radiobiological reactions that have a true threshold, the degree of manifestation of which depends on the absorbed dose (delayed cell division, acute radiation sickness or loss of organ function, for example) are classified by the ICRP as deterministic [1]. Thus, only in relation to deterministic reactions it makes sense to speak of "small doses", meaning subthreshold doses by them.

*Quasi-background doses*. Regardless the nature of the effect induced by ionizing radiation, i.e. regardless of whether this is stochastic or deterministic, from our point of view, most of the doses received by biological objects in the territories contaminated after the Chernobyl accident should be called "quasi-background doses", emphasizing their quantitative and qualitative similarity to the pre-accident parameters of the natural radiation background (NRF). Thus, the concept of "quasi-background dose" is closer in the meaning to the concept of "exposure dose" and characterizes to a greater extent the conditions of irradiation of a biological object, regardless of the thresholds of its sensitivity or stability. In particular, this concept can be useful in studying the nonlinear dependence of the frequency of the output of stochastic effects (aberrations, for example) in the region of doses close to the background values.

 The vertical distribution of radiocesium in the main root of an aquatic culture of seedlings as an example of the conditions for the formation of heterogeneous dose loads. Previously we studied the growth reaction of the roots of pea seedlings grown in an aqueous solution of chloride salt of cesium-137 [3]. At the specific activity of the used solutions from 1480 to 2220 *kBq/l* after a week of incubation of the plants, a complete cessation of the growth of their roots was observed. The calculation of the doses absorbed by the roots [4, 5] from external irradiation and exposure due to radionuclides incorporated into the root tissue was based on the assumption of a uniform distribution of radiocesium in the zones of the main root (meristematic zone, the zone of extension and differentiation, the zone of differentiated cells). The absorbed dose rate from internal exposure was determined by the formula:

 $P = 1,6 \times 10E^{-13} \times C \times f \times E$ 

where: *P* is the absorbed dose rate, *Gy/s*;

*C* is the specific activity of the sample, *Bq/kg*;

*f* is the output of this type of radiation to decay;

 *E* is the energy of particles (for beta particles - average), *MeV*.

The power of external irradiation in the zone of root habitation was 5 *mlR/h*. The value of the total absorbed dose accumulated during the time of plant incubation (7 days) was about 1 *Gy*, which in no way corresponded to the observed inhibitory (lethal) effect on the root meristem. Comparing the data of these experiments with the results of studying the effect of acute irradiation and knowing that the gamma and beta radiation of cesium-137 does not differ qualitatively from the standard radiation, we assumed the heterogeneity of the vertical distribution of radiocesium in the main root with its predominant concentration in the meristematic zone.

To test this assumption a series of experiments was carried out in which the vertical distribution of radiocesium along the length of the main root was studied. For this purpose after a day of incubation of 5-day seedlings of peas of the Zelenozerny variety and corn of the Dneprovskaya 247 variety in a radiocesium solution with a specific activity of 296 and 1700 *kBq/L*, respectively, the main root was cut along its entire length into 5-mm segments, weighed, and the total activity of each segment was measured on a 1211 RACKBETA liquid scintillation counter, after making sure that the self-absorption of beta radiation in the samples can be neglected [6].

The results of this series of experiments are shown in Figures 1. a, b**.** One can see a sharp uneven distribution of radiocesium along the length of the root. The first segment, in which the meristematic zone is located, has the highest specific activity. This was especially pronounced in maize roots. As the distance from the root tip, the activity of the segments drops sharply, and this drop has a noticeable oscillatory character. By incubating seedlings in a solution of radiocesium until lateral roots appeared and analyzing the distribution of radiocesium along the

length of the latter, we found that the distribution of radionuclides in them had a similar character (Figure 2.). Only some features of radiocesium accumulation by lateral roots should be noted. In particular, the total level of accumulation is much lower than in the main root, because by the time the lateral roots form, the main amount of the isotope (up to 98%) [7] is absorbed by the main root. In addition, some increase in the level of accumulation in the last segments of the lateral root was noted which is probably due to the secondary redistribution of the isotope in the main root.

Figure 3. presents the results of a more detailed study of the distribution of radiocesium within the first and the second segments with a total length of 10 mm. It can be seen that the main part of the activity is concentrated in the first three millimeters of the root apical zone. This is exactly the zone where the meristem is located, which consists of the most radiosensitive cells [8].

 Knowing the total activity of radiocesium in the main root, the weight of its individual segments, and the total activity of the isotope in each of them, it is possible to calculate what proportion of the total activity contains the apical part of the root and also to calculate the accumulation coefficients of the radionuclide by individual segments. It turned out that the apex 5 mm long whose weight is approximately 3% of the weight of the entire root (on the 5th-6th day of cultivation) contains up to 30% of the total activity of sorbed radiocesium. With an average accumulation coefficient for the entire root of about 800-1000, the accumulation coefficient in the apex reaches tens of thousands (27000), i.e. the concentration of radiocesium in the apical part and especially in the meristematic zone (accumulation factor 39000) is an order of magnitude higher than the average concentration of the radionuclide in the root, which means that the absorbed doses of gamma and beta radiation from incorporated radiocesium are at least 10 times higher than the initially established value, that is, it approaches a value of 10 *Gy*. This absorbed dose is already high enough to cause irreversible inhibition of growth processes. Figure 4. shows the dynamics of accumulation of radiocesium by the apical zone of the root, and in Figure 5. is shown the dynamics of the absorbed dose from external and internal exposure**.**

 The heterogeneity of the distribution of radiocesium established in this way confirms our assumption and makes it possible to explain the lethal effect of cesium radionuclides incorporated into the root tissue.

Based on data on the unequal intensity of biochemical processes along the length of the main root (respiration, protein synthesis, etc.) [6] and taking into account the chemical similarity of cesium with potassium, a series of experiments was carried out to elucidate the role of active and passive sorption of radiocesium in causing the registered heterogeneity of its accumulation. In the experimental version of the experiment, the roots of the seedlings were subjected to hyperthermic treatment before planting on the radiocesium solution by heating them in a water bath for 30 seconds at 50  $^{\circ}$ C. The results of this experiment (Figure 6.) show that, at least in the apical part of the root, the recorded level of radiocesium absorption is a consequence of active physiological and biochemical processes. However, these results cannot be considered definitive proof of the role of active sorption, since hyperthermia could also disrupt the processes of passive sorption.

In addition to experiments with cesium-137, similar studies were carried out with strontium-

90. An aqueous solution of strontium-90 nitrate was used. Without going into the methodological details of experiments with radiostrontium, we only note that a lethal effect similar to that observed for radiocesium was obtained at specific activities of radiostrontium that were three orders of magnitude higher than the corresponding values for radiocesium [3]. We found the explanation of this fact by analyzing the distribution of radiostrontium along the main root of pea seedlings. It can be seen from Figure 7**.** that radiostrontium is concentrated mainly in the zone of elongation and the beginning of cell differentiation which ultimately leads to significantly lower dose loads from beta radiation on the meristematic zone of the apical part of the root.

 In order to make sure whether the established features of the distribution of radionuclides over root zones are significant for the root systems of plants growing under soil culture conditions, it was decided to conduct a series of experiments on growing pea seedlings on soil contaminated with cesium-137 radionuclides. Soil with a specific activity for caesium-137 of 15 *kBq/kg* was used. Seedlings were removed from the soil from time to time and their main roots were cut into segments and analyzed by the method described above. The results presented in Figure 8. show a similar nature of the distribution of radionuclides along the length of the main root both in the water culture of seedlings and in the soil culture. The accumulation coefficient of radiocesium in the apical part of the root for 72 hours of incubation under these conditions was 200, and the specific activity was 3.5 *MBq/kg*. During the same period (comparatively short compared to the maximum possible duration of the growing season), the dose accumulated by the apical part under the conditions of soil culture was 0.02 *Gy* (in solution for the same period - 9.2 Gy). This circumstance once again indicates the important role of heterogeneity in the accumulation of radionuclides by different biological structures and the need to take it into account when estimating the doses absorbed by them, which may turn out to be very far from "low doses".

 The heterotropy of biological structures, revealed by studying the intake of radionuclides into them, causes the formation of heterogeneity of the doses absorbed by different structures, which can (at a high level of tropism) cause inhibition effects and even lethal effects. This circumstance is of fundamental importance for explaining radiobiological effects in plants growing in areas with an increased radiation background (zones of radionuclide anomalies). In particular, the most likely cause of the stimulatory effect of radiation on plants under such conditions may be a significant inhibition of proliferative activity or even irreversible damage to the root apical meristem which, as a rule, is followed by the removal of apical dominance, increased development of lateral roots resulting in the stimulation of the development of the aerial parts of plants .

 Thus, the detected vertical heterogeneity of the distribution of radiocesium in the main root of seedlings (with its predominant concentration in the meristematic zone) makes it possible to significantly refine the amount of doses absorbed by the root meristem from incorporated radionuclides, explaining the apparent discrepancy between the calculated absorbed doses and the observed effects. In addition, the established pattern of distribution of radiocesium in the root can serve as an experimental basis for developing a method for predicting radionuclide contamination of plants by analyzing the content of radionuclides in the root apexes.



**Figure 1. Specific activity of cesium-137 in 5-mm segments of the main roots of corn and pea seedlings, % of the activity of the apical segment**



**Figure 1.a. Specific activity of cesium-137 in 5-mm segments of the main roots of corn and pea seedlings, MBq/kg**



**Figure 1.b. Distribution of the total activity of cesium-137 over 5-mm segments of the main roots of corn and pea seedlings, Bq**



**Figure 2. Specific activity of caesium-137 in 5-mm segments of lateral roots of pea seedlings, % of the activity of the apical segment**



**Figure 3. Specific activity of cesium-137 in root segments depending on their distance from the apex of the main root of pea seedlings grown on an aqueous solution of cesium-137 chloride with a specific activity of 1.7 MBq/l, MBq/kg**



**Figure 4. Dynamics of accumulation of cesium-137 by the apical part of the main root and dynamics of growth of the main root of pea seedlings grown on a solution of cesium-137 chloride with a specific activity of 1.7 MBq/l, % to the corresponding indicator for the first day**



**Figure 5. Dose absorbed in the 5-mm apex of the main root of pea seedlings grown on an aqueous solution of cesium-137 chloride with an initial specific activity of 1.7 MBq/l, Gy**



**Figure 6. Specific activity of cesium-137 in 5-mm segments of the main root of pea seedlings after their hyperthermal treatment, MBq/kg**



**Figure 7. Specific activity of Sr-90 in segments of the main root of pea seedlings, % of the specific activity of the apical segment**



## **Figure 8. Specific activity in the root segments of pea seedlings grown on an aqueous solution of Cs-137 and on soil contaminated with a mixture of radionuclides of Chernobyl origin, % of the activity of the apical segment**

## **REFERENCES**

- [1] Radiation safety (1992). ICRP Recommendations 1990. ICRP Publication 60. Part 1. M.: *Energoatomizdat*, 192 p. (in Russian).
- [2] Akoev I. G. (1970). The problems of post-radiation recovery. M.: *Atomizdat*, 368 p. (in Russian).
- [3] Grodzinsky D. M., Kolomiets K. D., Kutlakhmedov Yu.A. (1991). Anthropogenic radionuclide anomaly and plants*.* Kyiv: Lybid, 92 p. (in Russian).
- [4] Grodzinsky D. M. (1962). Methodology for the use of radioactive isotopes in radiobiology. Kyiv: *Izdatelstvo akademii selskohozyaystvennyih nauk*, 172 p. (in Russian).
- [5] Moiseev A. A., Ivonov V. I. (1990). The Handbook of Dosimetry and Radiation Hygiene. M.: *Energoatomizdat*, 252 p. (in Russian).
- [6] Kozlov V. F. (1991). Radiation Safety Handbook. M.: *Energoatomizdat*, 352 p. (in Russian).
- [7] Mikheev A. N., Zezina N. V., Kutlakhmedov Yu. А., et al. (1991). New phenomena and approaches in investigation of soil-to-plant radionuclide transfer. In "Proceeding of I.U.R. Soviet Branch Seminar on the Radioecology and Counter-measures" Kyiv, DOC UIR, 209–224.
- [8] Gudkov I. N. (1980). The dynamics of the meristem and its radioresistance. In book. "Forms of post-radiation restoration of plants". Kyiv: *Naukova Dumka*, 82–115 (in Russian).
- [9] Kolosov I. I. (1962). The absorption activity of root systems. M.: *Izdatelstvo Akademii nauk SSSR*, 388 p. (in Russian).