Cytogenetic Effects of Radiation on Agricultural Plants Observed in the Chernobyl region during the First Years after the Accident

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Abstract

The cytogenetic consequences of radioactive contamination by the fallout after the accident at the Chernobyl NPP in 1986 to agricultural crops have been studied. In the acute period after the accident (1986), when the absorbed dose was mostly provided with external β- and γ-radiation, the radiation injury of agricultural crops resembled the effect produced by acute γ-radiation at comparable doses as basic cytogenetic tests testify. The yield of cytogenetic damage in leaf meristem of plants grown in the 10-km zone of the ChNPP in 1987-1989 (the period of chronic, lower level radiation exposure) was shown to be enhanced and dependent on the level of radioactive contamination. The rate of decline with time in cytogenetic damage induced by chronic exposure lagged considerably behind that of the radiation exposure. Analysis of genetic variability in three sequential generations of rye and wheat revealed increased cytogenetic damage in plants exposed to chronic radiation during the 2nd and the 3rd years.

Introduction

The accident at the Chernobyl NPP in 1986 is unique in both the extent of radioactive contamination and the values of the doses absorbed by living organisms. An estimation of total release of fission products (without noble gases) amounted to $1.85 \cdot 10^{18}$ Bq [1]. The areas most heavily exposed to radiation were natural and agricultural ecosystems within the 30-km ChNPP zone, with radioactive contamination reaching several thousands of MBq m^{-2} in 1986. The Chernobyl accident happened in late April - early May, i.e., the period of accelerated growth and formation of reproductive organs of plants, when their radiosensitivity is high. The maximum impact to living organisms was caused during 10-20 days after the accident when short-lived isotopes made a considerable contribution to the absorbed dose. During the summer and early autumn of 1986 the dose rate on the soil surface dropped to 20-25% of the initial value [2]. Since the autumn of 1986, the radiological situation in the exclusion zone has been stabilised; the acute irradiation at relatively high dose rates was replaced with low dose rate chronic exposure that persists up to the present time.

Cytogenetic and biological effects have been studied in rye seeds collected from agricultural fields in the 30-km ChNPP zone in 1986, and on intercalary leaf meristem of winter and spring crops grown on experimental plots in the 10-km ChNPP zone in 1987-1989.

Description of experimental plots

Seeds of winter rye of the "Belta" cultivar (from 50-100 main ears per plot) were collected on 4-6 August, 1986 from 5 m^2 plots in five fields with different dose rates of γ-radiation in the 30-km ChNPP zone. Seeds from a granary were used as control. The radiological characteristics of the experimental plots at the time of seed sampling are presented in Table 1.

The experimental plots in 1987-1989 were chosen with regard to the level of radioactive contamination of the territory, landscape and economic use of the land prior to the accident. Two pairs of

Table 1. Radiological characteristics of farm fields in the 30-km zone of the ChNPP.

Plot	σ, (MBq.m^{-2})	D_γ, (Gy)	D_β, (Gy)	$D_{\beta+\gamma}$, (Gy)
1. Kozhushki	19.6	0.18	1.15	1.33
2. Chamkov	58.1	0.33	2.80	3.13
3. Radin	61.1	0.58	3.40	3.98
4. Borzhshevka	129.5	1.20	6.50	7.70
5. Krasno	263.8	1.15	10.80	11.95

σ - total contamination density within 0-2 cm soil layer in August, 1986;
D_γ, D_β, $D_{\beta+\gamma}$ - doses absorbed over the vegetative period by plant critical organs.

Table 2. Radionuclide composition and contamination density on experimental plots in the 10-km zone of the ChNPP (May 15, 1988).

Plot	Location	Type of soil	Contamination density, (MBq.m^{-2})	Radionuclide composition, (%)				
				^{90}Sr	^{106}Ru	^{134}Cs	^{137}Cs	^{144}Ce
1	Meadow behind Chistogalovka	soddy podzolic	11.7	11.2	15.3	4.3	18.1	51.1
2	Meadow before Chistogalovka	sandy loam	106.0	8.1	13.7	5.3	25.0	47.9
3	ABZ	peaty humus gley	65.8	2.8	20.0	4.6	20.4	52.2
4	Red forest	peaty humus gley	454.0	9.1	13.1	4.6	20.7	52.5

plots were chosen to obtain comparative results. The plots in each pair were similar in soil type, but had diverse levels of radioactive contamination (Table 2). All four plots are situated 5-10 km west of the ChNPP along the Yanov-Chistogalovka road. The soils are typical for the Ukrainian Polesie, i.e. soddy podzolic, sandy and sandy loam, with different degrees of cultivation, a low availability of mineral nutrition and low sorption capacity for the most radionuclides. The arable layer has a low content of organic matter, available phosphorus and potassium, and a relatively high content of readily hydrolysable nitrogen. The soil solution reaction is acid (pH=4.4 - 4.6) and sub-acid (pH=5.1 - 5.3). A detailed description of plots was given in [3]. Winter (rye and wheat) and spring (barley and oats) crops were sowed using appropriate agricultural practices. The area of the experimental plots varied from 5 to 25 m^2. Samples of the leaf intercalary meristem of spring barley and oats were collected when the rudimentary ears reached 5 cm above the ground (at 2-3 weeks after sowing). Samples of winter rye and wheat were picked 2 times per year, at 3-4 weeks after sowing in the autumn and on the 10th day after the commencement of growth in spring

Materials and methods

Experiment in 1986

Seeds of winter rye collected in 1986 were allowed to germinate at 25-26° C. 15-30 seedlings were fixed at 16, 18, 20, 22, 24, 40, 48 and 72 hours of growth. Temporary squash slides prepared from the root apical meristem of each seedling were stained with aceto-orcein. In each slide, the mitotic index was calculated per 1000 cells, all ana-telophase cells were scored for aberrations; the aberration spectrum consisted of chromatid (single) and chromosome (double) bridges and fragments, as well as lagging chromosomes. 20-40 seedlings were simultaneously measured to determine the main root length. The laboratory germination rate was studied using the accelerated method of Stepanov [4].

To estimate the total density of radioactive contamination and the doses absorbed by the plants, soil was sampled by “envelop” method (taking samples from four corners and from the centre of the plot and calculating the density as an average) on each of the five fields. The samples were analysed by γ-

spectrometry of the upper layer (0-2 cm) samples.

The estimation of the total dose for rye over the vegetative period in 1986 was based on the radionuclide composition of the fallout and models describing both the growth of cereal plants and the radionuclide migration in the soil profile. The absorbed dose of γ-radiation was calculated by summing up the contributions from all lines of the spectrum obtained by γ-spectrometry on May 8, 1986 at assumption of a uniform radionuclide distribution in the soil to a depth of 1 cm. In calculations of the absorbed dose from β-radiation, the total β-radioactivity was assumed to consist of two parts. The first one is distributed over the soil surface and decreases from 100 % to 0 during the whole vegetative period (100 days) and the second one is distributed uniformly within the surface 1 cm soil layer and increases from 0 to 100 %. The absorbed doses of β-radiation were calculated by integrating the point source dose attenuation function for each radionuclide as described by [5]. The absorption coefficient for tissues covering the critical organ was set equal to 0.5.

Experiments in 1987-1989

Samples of the leaf intercalary meristem collected from the crop plants in 1987-1989 were fixed in acetic alcohol (1:3), stained with aceto-orcein and examined for aberrations (bridges and fragments) in ana-telophase. Five replicates were made for each plot at every sampling. Then, 100 ana-telophases were scored for each replicate.

An estimate of doses absorbed by the leaf meristem of plants in 1987-1989 was based on the data on soil radioactivity level and composition of the radionuclide contamination. Models of the infinite uniform source and of the infinite 20-cm thick layer source were used to calculate doses from β- and γ-radiation, respectively. The duration of the radiation exposure before sampling leaf meristem in spring crops was taken to be 30 days; in winter crops - 35 days for the autumn sampling and 210 days for sampling in the spring of the next year. When doses absorbed by a growing point over the whole vegetative period were estimated, the time between appearance of the growth point above the soil surface and harvesting was assumed to be equal to 100 days. γ-Radiation made the main contribution to absorbed dose during this time.

The radiometric analysis showed that the contamination density of soil varied considerably even within the area of one plot although the relative contents of radionuclides in all samples were similar (Table 2). Therefore, an average value for the density of radioactive contamination was calculated from the data on the absorbed dose rate in air. The ratio between the contamination density of soil and the absorbed dose rate in air was determined experimentally at the plot, which had the least variability in the measured levels of radioactive contamination. The dose rate of γ-radiation in air was measured with lithium fluoride thermoluminiscent dosimeters (TLD-100). Prior to measurements, the dosimeters were calibrated in a γ-radiation field of ^{137}Cs against the reference dosimeter 27012. To record γ-radiation only, the detectors were exposed in a 3 mm thick protective aluminium capsule. Changes in the dose rate of γ-rays with time were registered by DRG-01T dosimeter.

Optimum sample sizes needed for the estimation of examined values with a certain relative probable error at a given confidence level were determined by a method of statistical analysis of empirical distribution [6]. Student's test and confidence intervals were used to confirm statistical significance.

Results and Discussion

Winter rye, 1986

The experimental plots differed in the radioactive contamination of the soil surface layer and the dose absorbed by plant critical organs over the vegetative period by almost an order of magnitude (Table 1). According to the classification of zones of chronic radiation exposure of natural populations [7], the

Table 3. Aberrant cells in seedlings root meristems of winter rye seeds collected in the 30 km ChNPP zone (mean ± SE).

Plot	Dose, (Gy)	Cells in ana-telophase	Aberrant cells	Aberrant cell frequency, (%)
-	Control	952	40	4.20 ± 0.65
1	1.3	749	33	4.41 ± 0.75
2	3.1	1402	101	7.20 ± 0.69*
3	4.0	787	51	6.48 ± 0.88
4	7.7	447	49	10.96 ± 1.48**
5	12.0	668	95	14.22 ± 1.35***

Significance of variation from control: * - p<5%, ** - p<1%, *** p<0.1%;

Table 4. Damage distribution and severity of damage to aberrant cells in winter rye.

Plot	Dose, (Gy)	AC	AC_c	Number of cells with the following quantity of aberrations					Severity of damage, (aberrations per damaged cell)	MD, (%)
				1	2	3	4	5		
-	Control	40	4	27	5	4	0	0	1.36 ± 0.11	32.5
1	1.3	33	3	29	1	0	0	0	1.03 ± 0.10	12.1
2	3.1	101	9	64	23	3	1	1	1.39 ± 0.07	36.6
3	4.0	51	5	31	11	2	1	1	1.48 ± 0.19	39.2
4	7.7	49	5	29	9	4	2	0	1.52 ± 0.13	40.8
5	12.0	95	11	43	30	5	4	2	1.71 ± 0.12	54.7

Cells with complex indistinguishable damage were excluded from calculation of severity of damage.
AC - total number of aberrant cells; AC_c - number of cells with complex damage; MD is a percent of cells with multiple and complex damages.

radiation-induced biological effects should dominate over those of other ecological factors at least on the two experimental plots with the highest levels of radioactive contamination. The contribution of β-radiation to the total absorbed dose is different for each plot, but in all cases it is 6-9 times higher than that of γ-radiation.

Table 3 presents the results of the cytogenetic analysis of seedlings root meristems of winter rye seeds collected on contaminated plots. Even a dose of 3.1 Gy absorbed by plants results in a significant increase in the yield of aberrant cells. It is pertinent to note that significant increase in the aberrant cells rate in the root meristem of other agricultural crops was observed after exposure of seeds to γ-rays at comparable doses [8, 9].

Data on the damage distribution per cell are given in Table 4. To calculate the severity of damage in the aberrant cells, we used information on biological variability of this value among seedlings. The analysis of changes in the severity versus the absorbed dose showed a significant growth in the degree of cell damage along with radiation exposure. Overall, the severity of damage at chronic irradiation is within the limits registered in our previous experiments on exposure of seeds of agricultural crops to γ-rays at comparable doses [8,9].

Analysis of data on specific types of structural aberrations and their partial frequency (Table 5) has shown that there are no essential changes in the aberration spectrum with dose. The prevalent types of cytogenetic disturbance in the variety of rye examined, both for control and at chronic irradiation, are chromatid mutations followed by lagging chromosomes and chromosome aberrations. The low relative contribution of chromosome aberrations is supposed to be a specific feature of the biological material used. In our previous investigations [8,9] of structural mutations induction in root meristems of wheat and barley seeds exposed to ionising radiation, the contribution of chromosome aberrations was also small.

Table 5. Structural mutations spectrum in winter rye exposed to radioactive contamination.

Plot	Dose, (Gy)	TA	Different type aberrations									
			g	f'	m'	f''	m''	g	f'	m'	f''	m''
			Number					%				
-	control	49	5	27	13	1	3	10.2	55.1	26.5	2.0	6.1
1	1.3	31	4	18	9	0	0	12.9	58.1	29.0	0	0
2	3.1	128	9	63	41	5	10	7.1	49.2	32.0	3.9	7.8
3	4.0	68	13	32	18	0	5	19.1	47.0	26.5	0	7.4
4	7.7	67	7	34	23	0	3	10.4	50.8	34.3	0	4.5
5	12.0	144	19	59	55	4	7	13.2	41.0	38.2	2.8	4.9

Cells with complex indistinguishable damage are excluded.

TA - total number of aberrations; g - lagging chromosomes (genome mutations); f', m' - chromatid (single) fragments and bridges with associated fragments; f'', m'' - chromosome (double) fragments and bridges with associated fragments.

Table 6. Some viability indicators of seeds collected in the 30 km ChNPP zone (mean ± SE).

Plot	Dose, (Gy)	Mitotic index, (%)			Main root length, (mm)			Germination,(%)
		16-24 h	40-48 h	72 h	16-24 h	40-48 h	72 h	
-	Control	1.69 ± 0.25	3.83 ± 0.26	3.09 ± 0.17	1.96 ± 0.19	22.95 ± 1.19	34.21 ± 3.42	62.0 ± 2.4
1	1.3	1.16 ± 0.23	2.64 ± 0.23*	3.24 ± 0.79	2.22 ± 0.20	12.85 ± 1.13	34.46 ± 2.17	87.5 ± 1.7*
2	3.1	1.73 ± 0.22	2.29 ± 0.17*	3.95 ± 0.35	2.27 ± 0.17	17.75 ± 0.97	37.97 ± 2.93	74.5 ± 2.2*
3	4.0	1.25 ± 0.18	2.54 ± 0.18*	3.79 ± 0.26	1.83 ± 0.14	13.35 ± 0.93	45.29 ± 3.33	88.8 ± 1.6*
4	7.7	0.83 ± 0.17	2.75 ± 0.16*	3.99 ± 0.25	1.64 ± 0.11	12.70 ± 0.77	36.06 ± 3.21	70.8 ± 2.3
5	12.0	1.11 ± 0.20	2.61 ± 0.27*	2.98 ± 0.32	1.93 ± 0.24	13.80 ± 0.73	25.48 ± 3.49	31.2 ± 2.3*

Mean values over the first (16-24 h) and the second (40-48 h) mitosis are presented.
Significance: * - $p<5\%$.

Such an unusual result may be due to the possible induction at G_1 stage, of long-lived potential damage that is later realised as genuine mutations at S and G_2 stages. Dubinin & Nemtsova [10] have obtained similar results analysing aberration spectrum in root meristems of irradiated seeds of agricultural plants.

Table 6 presents data on some viability indicators of seeds collected on the plots in the 30 km ChNPP zone. The mitotic indices determined at 8 time points (16-72 hours after germination) show that there is an insignificant tendency for mitotic activity to decline with the increase in the dose absorbed. A significant radiation depression of mitotic activity was observed only in 40-48-hours seedlings. However, at this time point, the mitotic indices don't correlate with dose. Seed germinating power shows statistically significant stimulation effects at doses of 1.3-4.0 Gy, and considerable, statistically significant inhibition at the dose of 12 Gy. Attention should be paid to the fact that some of the morphological indices illustrating the viability of germs at early stages of ontogenesis depend on dose. It should also be noted that morphological stimulation (Table 6) was accompanied by high and significant increment of cytogenetic disturbances. This result appears a good illustration of the ambiguous nature of "stimulation" and indicates that the term should not be considered as a synonym for "benefit" or even harmlessness of exposure, because this outcome, assumed to be positive in terms of economic efficacy, may simply be a small part of a wider biological phenomenon that is negative overall.

Thus, in the first acute period after the Chernobyl accident, when the absorbed dose was primarily formed by external β- and γ-radiation, the radiation injury to agricultural crops, according to the basic cytogenetic tests, resembled the effect produced by acute γ-irradiation at comparable doses.

Table 7. Aberrant cell frequency in the leaf meristem of spring crops grown on experimental plots in the 10-km ChNPP zone.

Plot	Dose, (cGy)	Barley		Oats	
		Number of AC	Frequency of AC, (%)	Number of AC	Frequency of AC, (%)
1	2.05	142	28.4 ± 2.0	203	40.6 ± 2.2
3	12.09	181	36.2 ± 2.1	161	32.2 ± 2.1
2	17.90	156	31.2 ± 2.2	274	54.8 ± 2.2*
4	78.46	261	52.2 ± 2.2*	271	54.2 ± 2.2*

AC is aberrant cells. Significance of variation from plot 1: * - p<5%.

Spring barley and oats, 1988

Experiments with spring crops (barley and oats) were only carried out in 1988. Table 7 gives the data on the frequency of aberrant cells in the leaf meristem of barley and oats grown on plots with a nearly 40-fold difference in radioactive contamination density. The yield of aberrant cells trends to rise with the absorbed dose, indicating the radiation-induced nature of the observed changes. The levels of cytogenetical damage in oats generally exceed those in barley at the same absorbed doses, which is the reverse of the relationship for values of LD_{50} reported by [11].

In this study there are unexpectedly high levels of cytogenetic disturbances in comparison with our previous experiments on acute [12,13] and chronic γ-irradiation of barley plants [14]. According to [12-14], the spontaneous level of aberrant cells in the leaf meristem of barley was 10-14%. In the present study, the yield of cytogenetic disturbances was 2-3 times higher than the spontaneous occurrence expected from our works on acute γ-irradiation of barley plants even at plots with the lowest levels of radioactive contamination. Such an apparent discrepancy between the level of genetic effects expected from the dosimetric data and the level observed in the experiment has also been reported by other authors who had carried out investigations in the 30-km Chernobyl zone [15,16].

Winter wheat and rye, 1987-1989

An estimation of consequences of radioactive contamination should take into consideration that winter crops are subjected to radiation for a longer time and their leaf meristem lies within the upper, most densely contaminated soil layer during the winter dormancy period. Experiments with winter crops (wheat and rye) were carried out from the autumn of 1987 till the autumn of 1989. Therefore, from the data available, conclusions can be drawn not only about dose dependence of cytogenetic damage rate but also about how the plant response to radiation changed as the absorbed dose decreased with time due to both radioactive decay and the migration of radionuclides in soil.

Frequencies of aberrant cells in the leaf meristem of winter rye and wheat grown on plots with diverse levels of radioactive contamination are given in Table 8. For winter wheat, there is a significant decrease in the yield of cytogenetic disturbances with time from the accident at all plots except plot 3. For winter rye, such a decrease is observed at plots 2 and 4. At the same time, the yields of the aberrant cells per dose unit rise with time in all autumn samplings (Table 8). This means that the rate of decline in the cytogenetic damage to plants lags behind that of radiation exposure. This phenomenon was also reported by [17].

The regression analysis for each year and for each sampling time has shown a general significant development in the frequency of aberrant cells with increase in the dose absorbed by plants, indicating thereby the role of radiation exposure in the observed alterations. However, no significant dose dependence was revealed: a) in the autumn of 1989, in rye ($p<0.07$), when the absorbed dose decreased by 2.6 times compared with 1987 and b) in the spring of 1988, in wheat ($p<0.22$) and rye ($p<0.06$), when doses absorbed by plants were maximal over the investigated period and one could expect an apparent dose-effect relationship. If we distinguish two sets of the data for peaty humus gley (plots 3 and 4) and for soddy podzolic sandy loam (plots 1 and 2) soils, there is the expected dose dependence, as seen in the case

Table 8. Aberrant cells frequency in leaf meristem cells of winter crops grown in the 10-km ChNPP zone.

Sampling time	Dose, (cGy)	Rye		Wheat	
		AC, %	AC per 1 cGy	AC, %	AC per 1 cGy
Plot 1 (soddy podzolic soil)					
Autumn 1987	3.88	19.0 ± 1.8	4.90 ± 0.97	32.4 ± 2.1	8.35 ± 1.24
Autumn 1988	1.99	21.4 ± 1.8	10.75 ± 1.39	23.0 ± 1.9 [a]	11.56 ± 1.43
Autumn 1989	1.03	20.2 ± 1.8	19.61 ± 1.78	18.0 ± 1.7 [a]	17.48 ± 1.70
Spring 1988	17.94	26.0 ± 2.0	1.45 ± 0.53	53.2 ± 2.2	2.97 ± 0.76
Spring 1989	10.26	20.4 ± 1.8	1.99 ± 0.62	23.2 ± 1.9 [b]	2.26 ± 0.66
Plot 3 (peaty humus gley soil)					
Autumn 1987	23.15	20.2 ± 1.8	0.87 ± 0.42	24.0 ± 1.9	1.04 ± 0.45
Autumn 1988	11.67	25.4 ± 1.9	2.18 ± 0.65	24.8 ± 1.9	2.13 ± 0.64
Autumn 1989	5.81	23.4 ± 1.9	4.03 ± 0.88[a]	21.2 ± 1.8	3.65 ± 0.84[a]
Spring 1988	106.50	32.8 ± 2.1	0.31 ± 0.25	34.6 ± 2.1	0.32 ± 0.25
Spring 1989	59.74	22.0 ± 1.9 [b]	0.37 ± 0.27	24.0 ± 1.9 [b]	0.40 ± 0.28
Plot 2 (sandy loam soil)					
Autumn 1987	33.50	32.4 ± 2.1	0.97 ± 0.44	33.4 ± 2.1	1.00 ± 0.44
Autumn 1988	17.48	24.2 ± 1.9 [a]	1.38 ± 0.52	25.6 ± 2.0	1.46 ± 0.54
Autumn 1989	9.36	20.6 ± 1.8 [a]	2.20 ± 0.66	20.6 ± 1.8 [a]	2.20 ± 0.66
Spring 1988	155.75	59.6 ± 2.2	0.38 ± 0.28	68.6 ± 2.1	0.44 ± 0.30
Spring 1989	90.63	24.6 ± 1.9 [b]	0.27 ± 0.23	24.6 ± 1.9 [b]	0.27 ± 0.23
Plot 4 (peaty humus gley soil)					
Autumn 1987	148.78	45.2 ± 2.2	0.30 ± 0.25	39.6 ± 2.2	0.27 ± 0.23
Autumn 1988	76.14	26.6 ± 2.0 [a]	0.35 ± 0.26	27.6 ± 2.0 [a]	0.36 ± 0.27
Autumn 1989	39.60	22.8 ± 1.9 [a]	0.58 ± 0.34	25.8 ± 2.0 [a]	0.65 ± 0.36
Spring 1988	687.20	47.4 ± 2.2	0.07 ± 0.12	42.8 ± 2.2	0.06 ± 0.11
Spring 1989	392.49	29.8 ± 2.0 [b]	0.08 ± 0.12	29.8 ± 2.0[b]	0.08 ± 0.12

AC – frequency of aberrant cells;
[a] and [b] – difference from the level of the autumn 1987 and the spring 1988, correspondingly, is significant, $p<0.05$.

of the spring crops. Attention is drawn to the abnormally low level of cytogenetic disturbances in wheat at plot 3 in the autumn of 1987 and spring of 1988, which differ significantly from that at plot 1 with the lowest level of radioactive contamination. The reason for this abnormal observation is not clear yet.

It is interesting that slopes of fitted linear dose curves in autumn are considerably higher than in spring of the following year for both cereals. So, the yield of aberrant cells in autumn has closer correlation with dose absorbed by plants than that in spring. Dose was accumulated over a prolonged time in spring samples, so cytogenetic damage probably depends not as much on radiation as on other factors.

Variability in the plant generations

Over the entire period of investigations (autumn of 1987 - autumn of 1989), a part of the seeds collected was planted again on the same plots, allowing study of genetic variability in three successive generations of winter rye and wheat. The doses absorbed by a growing point of plant over the whole vegetative periods from planting to harvesting of 1987-1988 and 1988-1989 were in the range of 18 - 717 cGy and 11 - 418 cGy, respectively. Each of these doses accumulated during vegetation is a sum of two values, i.e. the dose absorbed from planting in autumn to the time of spring sampling (Table 8) and a dose absorbed from spring sampling to seed harvesting. The last value is comparatively small because the most part of dose is delivered to plants with β-radiation while the growing point is under or near the soil surface prior to the spring sampling.

In rye, in the autumn of 1989, at plots 2 and 4 with the highest level of radioactive contamination, the

frequencies of aberrant cells in the leaf meristem in plants growing on contaminated sites in the second year (marked as the X_2 generation) and the third year (the X_3 generation) significantly exceed the values for those growing only in the first year (the X_1 generation) (Table 9).

In contrast to the results obtained by [18], the level of genetic variability in the X_3 generation in the present study does not decline and even somewhat increases (Figure 1), though there is no statistically significant difference between the yields of aberrant cells in the X_2 and X_3 generations. The results of our study correspond well with the data reported by [15] who observed a rise in mutation load in the *Arabidopsis thaliana* populations at all levels of radioactive contamination during the first 2-3 years after the accident. In wheat, this tendency is more pronounced. In the autumn of 1989, a significant increase in the frequencies of aberrant cells in the X_2 and X_3 generations, compared with X_1, was recorded at 3 out of 4 plots (Table 9). Differences between the numbers of aberrant cells in the X_2 and X_3 generations are small and statistically insignificant for both crops (Figure 1).

One of the possible explanations of the observed phenomenon is related to genome destabilisation in plants grown from seeds affected by radiation. Results from numerous experiments carried out with representatives of different kingdoms of the living world [17,19] indicate that chronic irradiation in regions affected as a result of the accident at the Chernobyl NPP can cause heritable destabilisation of genetic structures that appears, in particular, as an increased yield of cytogenetic disturbances and kariotypic variability in the offspring of irradiated organisms. From these viewpoints, the phenomenon observed in this study may be a reflection of the first stage in cytogenetic adaptation [20,21], that is,

Table 9. Aberrant cells frequency in leaf meristem of winter rye and winter wheat in 3 subsequent plant generations grown on the plots in the 10-km ChNPP zone (sampling time is autumn 1989).

Plot	D_{87-88}, (cGy)	D_{88-89}, (cGy)	D, (cGy)	Rye		Wheat	
				Number of aberrant cells	Aberrant cell frequency, (%)	Number of aberrant cells	Aberrant cell frequency, (%)
X_1-generation (first year of planting on contaminated plots)							
1			1.03	101	20.2 ± 1.8	90	18.0 ± 1.7
3			5.81	117	23.4 ± 1.9	106	21.2 ± 1.8
2			9.36	103	20.6 ± 1.8	103	20.6 ± 1.8
4			39.60	114	22.8 ± 1.9	129	25.8 ± 2.0
X_2-generation (second year of planting on contaminated plots)							
1		10.68	1.03	119	23.8 ± 1.9	130	26.0 ± 2.0*
3		63.54	5.81	144	28.8 ± 2.0	135	27.0 ± 2.0
2		95.93	9.36	155	31.0 ± 2.1*	146	29.2 ± 2.0*
4		417.49	39.60	165	33.0 ± 2.1*	170	34.0 ± 2.1*
X_3-generation (third year of planting on contaminated plots)							
1	18.45	10.68	1.03	136	27.2 ± 2.0	132	26.4 ± 2.0*
3	111.1	63.54	5.81	150	30.0 ± 2.0	157	31.4 ± 2.1*
2	162.05	95.93	9.36	179	35.8 ± 2.1*	150	30.0 ± 2.0*
4	717.2	417.49	39.60	163	32.6 ± 2.1*	167	33.4 ± 2.1

D – dose, accumulated from planting in autumn 1989 to the sampling time;

D_{87-88}, D_{88-89} – doses, accumulated by parent plants during the whole vegetative period from planting up to harvesting in 1987-1988 and 1988-1989 years, respectively

X_1-generation – plants grown from intact seeds and accumulated dose D from planting in autumn 1989 to the sampling time;

X_2-generation – parent plants were sown in 1988, harvested in summer 1989 and planted again on the same plots in autumn 1989. Genetical effects are the result of both the ancestral dose D_{88-89} and current exposure D;

X_3-generation - parent plants grew on the same plots in 1987-1988 and in 1988-1989 and accumulated doses D_{87-88} and D_{88-89}, correspondingly. Seeds harvested in 1989 were sown again in autumn 1989 and plants of the X_3 generation got dose D.

Significance of variation from the level of cytogenetic disturbances in the X_1 generation: * - $p<5\%$.

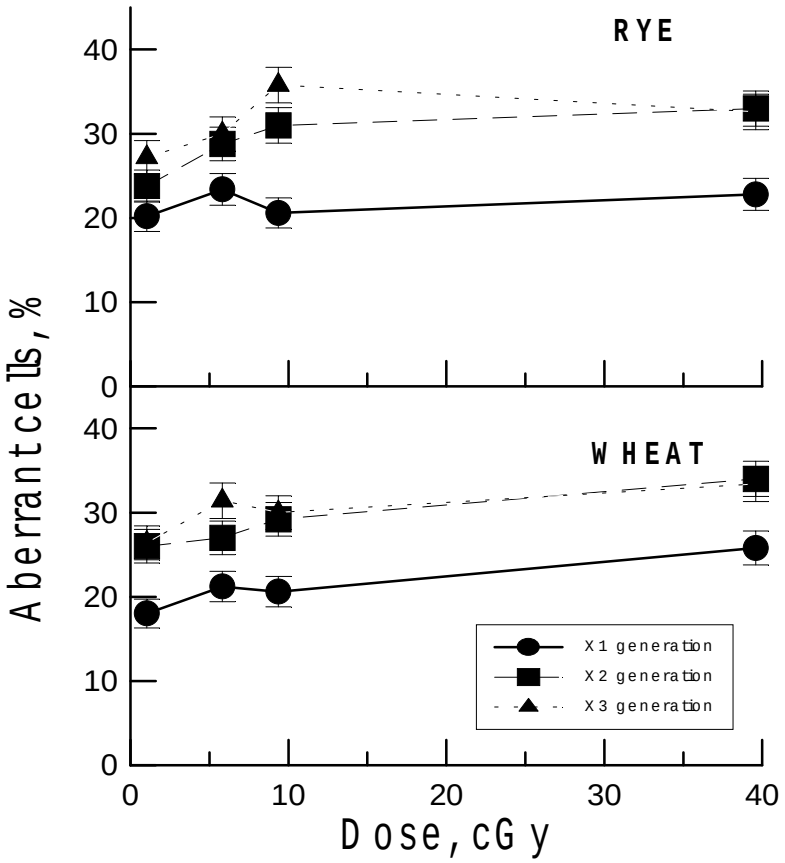

Figure 1. Yield of aberrant cells in three successive generations of winter rye and wheat, grown on contaminated plots.

chronic exposure to low-level radiation possibly alters the genetic structure of population. Response to an external impact, such as an expansion of the gene pool variability providing a potential basis for the subsequent selection of the most adaptable forms, is a reflection of the fundamental mechanisms (grounding the basis for life) [22] that ensure resistance of living systems and their possible adaptation to varying conditions of the environment.

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