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Journal of Environmental Radioactivity 66 (2003) 155–169
JOURNAL OF ENVIRONMENTAL RADIOACTIVITY
www.elsevier.com/locate/jenvrad

Genetic consequences of radioactive contamination by the Chernobyl fallout to agricultural crops

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Received 10 May 1999; received in revised form 26 February 2002; accepted 1 March 2002

Abstract

The genetic consequences of radioactive contamination by the fallout to agricultural crops after the accident at the Chernobyl NPP in 1986 have been studied. In the first, acute, period of this accident, when the absorbed dose was primarily due to external β- and γ-irradiation, the radiation injury of agricultural crops, according to the basic cytogenetic tests, resembled the effect produced by acute γ-irradiation at comparable doses. 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 irradiation during the 2nd and 3rd years.

Keywords: Chernobyl accident; Radioactive contamination; Chromosome aberrations; Agricultural crops; Genetic consequences

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0265-931X/03/$ - see front matter © 2003 Elsevier Science Ltd. All rights reserved.
doi:10.1016/S0265-931X(02)00121-2

1. 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. The Chernobyl release also has a broader set of radionuclides as compared with other accidents such as that in the South Urals in 1957. The complex time dependence of the individual radionuclide releases, and the variable meteorological conditions during the ten days of active emission of radioactive materials, resulted in an extremely heterogeneous fallout pattern. The non-uniform radionuclide distribution in the environment caused a wide variation in the total doses absorbed by living organisms; in a number of cases, this was greater than an order of magnitude even in small, localised areas.

The areas most exposed to radiation were natural and agricultural ecosystems within the 30-km ChNPP zone, with radioactive contamination over a significant part reaching several thousands of MBq m^{-2} in 1986. The Chernobyl accident took place 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 exposure of living organisms occurred within the period of acute irradiation (up to 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. Since the autumn of 1986, the radiological situation in the exclusion zone has been stabilised; the acute irradiation at relatively high dose rates has been 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 zone of the ChNPP in 1986 and on intercalary leaf meristem of winter and spring crops grown on experimental plots in the 10-km zone of the ChNPP in 1987–1989.

2. Characteristics 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 in the 30-km zone of the ChNPP from 5 m^2 plots in five fields with different dose rates of γ-radiation. Control seeds for the determination of spontaneous levels of cytogenetic damage were obtained from a granary. 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 by taking account of the level of radioactive contamination of the territory, the landscape and the economic use of the land prior to the accident. To obtain comparative results, two pairs of plots were chosen which were similar in soil type, but had different 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

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

Plot	σ^a, (MBq.m^{-2})	$D_\gamma{}^b$, (Gy)	$D_\beta{}^b$, (Gy)	$D_{\beta+\gamma}{}^b$, (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

[a] σ—total contamination density within 0–2 cm soil layer in August, 1986
[b] Dγ, Dβ, $D\beta_{+\gamma}$—doses absorbed over the vegetative period by the critical organs of the plant

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

Plot	Location	Type of soil	Contamination density	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

low availability of mineral nutritients and low sorption capacity for the majority of 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 has been given previously by Krouglov et al. (1997). Winter (rye and wheat) and spring (barley and oats) crops were sowed using appropriate agricultural practices. The areas of the experimental plots varied from 5 to 25 m^2. Samples of the leaf intercalary meristem of spring barley and oats were selected when the rudimentary ears reached 5 cm above the ground (2–3 weeks after sowing). Samples of winter rye and wheat were taken two times per year, at 3–4 weeks after seeding in the autumn and on the tenth day after the commencement of growth in spring.

3. Materials and methods

Seeds of winter rye collected in 1986 were allowed to germinate at 25–26° C. Fixation of 15–30 seedlings was carried out after 16, 18, 20, 22, 24, 40, 48 and 72

h of growth. Temporary squash slides for cytoanalysis were prepared from the root apical meristem of each seedling and stained with aceto-orcein. In each slide, the mitotic index was calculated per 1000 cells, all ana-telophase cells were scored to determine the fraction of cells with aberrations; the aberration spectrum analysis was performed with the separation of chromatid (single) and chromosome (double) bridges and fragments, as well as the identification of lagging chromosomes. In the assessment of lagging, only those chromosomes which lay at a distance of at least two times greater than the chromosome thickness from the separating anaphase caps were taken into account. Simultaneously, 20–40 seedlings were measured to determine the main root length. The laboratory germination rate was studied using the accelerated method of Stepanov (1969).

To estimate the total density of radioactive contamination and the doses absorbed by the plants, samples of soil were taken by 'the 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 assuming a uniform radionuclide distribution to a depth of 1 cm in the soil. In calculations of the absorbed dose from β-radiation, the total β-radioactivity was assumed to be distributed as two time-dependent sources. The first source is distributed over the soil surface and decreases from 100 to 0% of the deposited activity during the whole vegetative period (100 days), and this is re-distributed to form the second source uniformly within the surface 1 cm soil layer where the concentration increases from 0 to 100%. An angular distribution of β-radiation from the surface source was assumed to be cosinusoidal to take account of β-radiation screening by soil microrelief; an angular distribution from the 'thick layer' source was also assumed to be cosinusoidal because of the similarity between the cosinusoidal law and the diffuse distribution of β-radiation from the 'thick layer' source. The absorbed doses of β-radiation were calculated by integrating the point source dose attenuation function for each radionuclide as described by Spirin (1997). The absorption coefficient for tissues covering the critical organ was set equal to 0.5.

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

The doses absorbed by the leaf meristem of plants in 1987–1989 were estimated based on data on the radioactivity content of the soil and the radionuclide composition. 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 period 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 the appearance of the growth point above the soil surface and harvesting was assumed to be equal to 100 days. The main contribution to absorbed dose during this period was caused by γ-radiation.

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 absorbed 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. The DRG-01T dosimeter was used to determine changes in the absorbed dose rate of γ-radiation with time.

A method of statistical analysis of empirical distribution was used to determine an optimum sample size needed for the estimation of examined values with a certain relative probable error at a given confidence level (Geraskin et al., 1994). Student's test and the confidence intervals method were used to confirm statistical significance.

4. Results and discussion

4.1. Winter rye, 1986

Table 1 shows that 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. According to the classification of zones of chronic ionising radiation exposure of natural populations (Polikarpov and Tsytsugina, 1995), the 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. This is in good accordance with the results of dose estimation in experiments on contamination of agricultural crops by nuclear fission products (Prister et al., 1982).

Table 3 presents the results of the cytogenetic analysis of seedling root meristems of winter rye seeds collected on plots with different levels of radioactive contamination. Statistical analysis shows that 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, in our experimental studies on the induction of cytogenetic effects by acute γ-

Table 3
Change in the number of aberrant cells in the root meristem of winter rye seedlings with irradiation dose (mean±SE)[a]

Plot	Dose, (Gy)	Number of cells in ana-telophase	Number of 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∗∗∗

[a] Significance of variation from control: ∗=5%, ∗∗=1%, ∗∗∗=0.1%

irradiation of seeds of other agricultural crops (Geraskin et al., 1995, 1997), a significant increase in the aberrant cells rate in the root meristem was observed at comparable doses.

Data on the damage distribution per cell as a function of dose 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 with increasing radiation exposure. Overall, the severity of damage induced by chronic irradiation is within the limits we observed in experiments with acute γ-irradiation of seeds of agricultural crops at comparable doses (Geraskin et al., 1995, 1997).

Data on specific types of structural aberrations and their partial frequency as a

Table 4
Damage distribution per cell and severity of damage to aberrant cells in winter rye[a]

Plot	Dose, (Gy)	AC[b]	AC_c[b]	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

[a] The cells with complex damages that could not be resolved were excluded in calculation of severity of damage

[b] AC—total number of aberrant cells; AC_c—number of cells with complex damage; MD is a percent of damaged cells with multiple and complex damages

function of dose are presented in Table 5. Statistical analysis of the data presented 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 chronic irradiation conditions, are chromatid mutations followed by lagging chromosomes and chromosome aberrations. The low relative yield of chromosome aberrations is presumed to be a specific feature of the biological material used. In our previous investigations (Geraskin et al., 1995, 1997) of structural mutation induction in the root meristem of wheat and barley seeds exposed to ionising radiation, the contribution of chromosome aberrations was also small. Such an unusual result may be due to the possible induction, in the G_1 phase, of long-lived potential damage that is later realised as genuine mutations in phases S and G_2. Similar results were obtained by Dubinin and Nemtsova (1972) from an analysis of the aberration spectrum in the root meristem of irradiated seeds of agricultural plants.

Table 6 presents data on changes in some indicators of seed viability as a function of the total dose absorbed by plants. The mitotic index determined at 8 time points (16–72 h after germination) has shown 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 seedlings at 40–48 h after germination. However, at this time point, the mitotic index did not show a correlation with dose. The analysis of seed germinating power revealed statistically significant stimulation effects at doses of 1.3–4.0 Gy; at the dose of 12 Gy a considerable, and statistically significant, inhibition was observed. Attention should be paid to the fact that some of the morphometric indices showing the viability of germinating seeds at early stages of ontogenesis are connected with dose. It should be also noted that the morphometric stimulation (Table 6) was attended by a high and significant increment of cytogenetic disturbances (by a factor of 2.6 over the control value, Table 3). This result appears to be a good illustration of the ambiguous nature of ‘stimulation’ and indicates that the term should not be considered as a synonym for

Table 5
Characteristics of structural mutations spectrum versus dose absorbed by winter rye[a]

Plot	Dose, (Gy)	TA	Number of different type aberrations					Percent contribution of different types of aberrations				
			g	f’	m’	f”	m”	g	f’	m’	f”	m”
–	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

[a] The cells with complex damages that could not be resolved 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
Variations in some indicators showing seed viability with dose absorbed by winter rye (mean±SE)[a]

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∗

[a] Mean values over the first (16–24 h) and the second (40–48 h) mitosis are presented Significance: ∗=5%

'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 of the Chernobyl accident, when the absorbed dose was primarily formed from external β- and γ-irradiation, the radiation injury to agricultural crops, according to the basic cytogenetic tests, resembled the effects produced by acute γ-irradiation at comparable doses.

4.2. Spring barley and oats, 1988

Experiments with spring crops (barley and oats) were only carried out in 1988. Table 7 gives 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 tends to rise with the increase in the absorbed dose, indicating the radiation-induced nature of the observed changes. The levels of cytogenetic damage in oats tend to be greater than those in barley at the same absorbed doses, which is the reverse of the relationship for values of LD_{50} reported by Sarapultzev and Geraskin (1993).

In this study, an unexpectedly high level of cytogenetic disturbances occurred in comparison with the results of our previous experiments on acute (Geraskin et al., 1993; Geraskin et al., 1996) and chronic γ-irradiation of barley plants (unpublished data). According to our previous investigations (Geraskin et al., 1993; Geraskin et al., 1996), 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. This apparent discrepancy between the impacts implied by the dosimetric data and the observed cytogenetic effects has also been reported by other authors who have carried out investigations in the 30-km Chernobyl zone (Abramov et al., 1995; Al'ferovich et al., 1996).

Table 7
Aberrant cells frequency in leaf meristem cells of spring crops grown on experimental plots with different levels of radioactive contamination[a]

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∗

[a] AC is aberrant cells. Significance of variation from plot 1: ∗=5%

4.3. Winter wheat and rye, 1987–1989

An estimation of the consequences of radioactive contamination should take into consideration that winter crops are subjected to irradiation for a longer time and their leaf meristem lies within the upper, most 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 the rate of cytogenetic damage but also about how the plant response to irradiation changed as the absorbed dose decreased with time due to both radioactive decay and the migration of radionuclides in the soil.

Analysis of the data on the frequency of aberrant cells in the leaf meristem of winter rye and wheat (Table 8) grown on plots with various levels of radioactive

Table 8
Aberrant cells frequency in leaf meristem cells of winter crops on experimental plots with different levels of radioactive contamination

Plot	Dose, (cGy)	Rye		Wheat	
		AC[c], %	AC[c] per 1 cGy	AC[c], %	AC[c] 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

[a] difference from the level of the autumn 1987 is significant, $p < 0.05$
[b] difference from the level of the spring 1988 is significant, $p < 0.05$
[c] AC—frequency of aberrant cells

contamination shows that for winter wheat there is a significant decrease in the yield of cytogenetic disturbances with time elapsed from the accident at all plots except plot 3, and for winter rye such a decrease is seen for plots 2 and 4. At the same time, the yields of the aberrant cells per dose unit rise with time in all autumn sampling (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 Shevchenko et al. (1995).

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 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. As in the case of the spring crops, the expected dose dependence is apparent if we distinguish data for peaty humus gley (plots 3 and 4) and soddy podzolic sandy loam (plots 1 and 2) soil. Attention is drawn to the abnormally low level of cytogenetic disturbance in wheat at plot 3 in the autumn of 1987 and spring of 1988, which differed significantly from that in plot 1 with the lowest level of radioactive contamination. 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 the autumn had a closer correlation with the dose absorbed by plants than was observed in the spring. Dose was accumulated over a prolonged time for the spring samples, so cytogenetic damage probably depended not as much on the radiation as on other factors.

4.4. Variability in the plant generations

Over the entire period of investigations (autumn of 1987–autumn of 1989), a proportion of the seeds collected was planted again on the same plots, allowing an analysis to be made of the genetic variability in three successive, irradiated generations of winter rye and wheat. The doses absorbed by a growing point of a plant over the whole vegetative periods from planting to harvesting in 1987–1988 and 1988-1989 were in the range of 18–717 cGy and 11–418 cGy, respectively. Each of these doses accumulated during vegetative growth is a sum of two values, i.e., the dose absorbed from planting in autumn to the time of spring sampling (Table 8) and the dose absorbed from spring sampling to seed harvesting. The last value is comparatively small as the greater part of the dose is delivered to plants from β-radiation while the growing point is under, or near, the soil surface, i.e., prior to the spring sampling. In rye, in the autumn of 1989, the frequency of aberrant cells in the leaf meristem in plants growing on contaminated sites in the second (marked as the X_2 generation) and third years (the X_3 generation) significantly exceeded the values for those growing in the first year (the X_1 generation) at plots 2 and 4 with the highest level of radioactive contamination (Table 9). In contrast to the results

Table 9
Aberrant cells frequency in leaf meristem cells of winter rye and winter wheat in 3 subsequent plant generations grown on plots with different levels of radioactive contamination (sampling time is autumn 1989)[f]

Plot	$D_{87–88}$[b], (cGy)	$D_{88–89}$[b], (cGy)	D[a], (cGy)	Rye		Wheat	
				Number of aberrant cells	Aberrant cell frequency, (%)	Number of aberrant cells	Aberrant cell frequency, (%)
X_1[c]-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[d]-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[e]-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

[a] D—dose, accumulated from planting in autumn 1989 to the sampling time

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

[c] X_1-generation—plants grown from intact seeds and accumulated dose D from planting in autumn 1989 to the sampling time

[d] 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

[e] 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 received dose D

[f] Significance of variation from the level of cytogenetic disturbances in the X_1 generation: ∗=5%

obtained by Kalchenco et al. (1981), in the present study, the level of genetic variability in the X_3 generation does not decline and even somewhat increases (Fig. 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 Abramov et al. (1995) 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 frequency of aberrant cells in the X_2 and X_3 generations, compared with X_1, was recorded for 3 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 (Fig. 1).

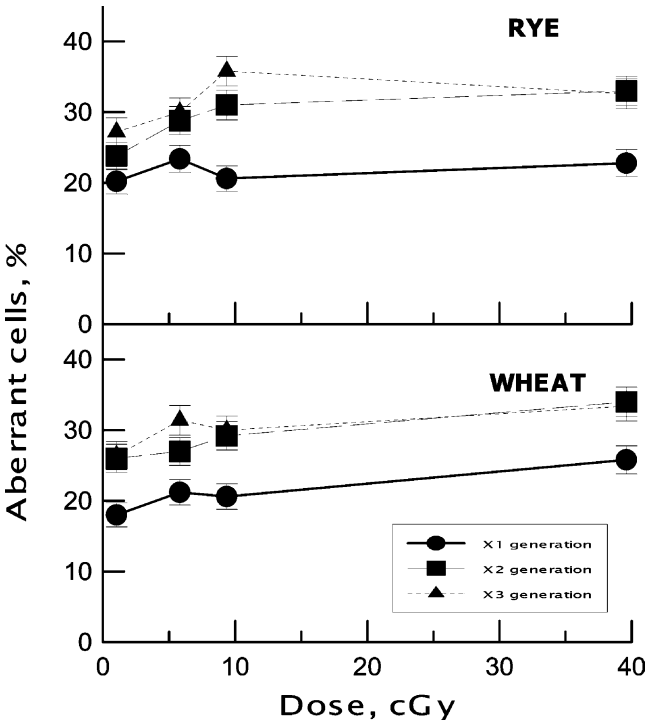

Fig. 1. Yield of aberrant cells in three successive generations of winter rye and wheat, grown on contaminated plots.

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 (Shevchenko et al., 1995; Pelevina et al., 1996) 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 disturbance and karyotypic variability in the offspring of irradiated organisms. From these viewpoints, the phenomenon observed in this study may be a reflection of the first stage of cytogenetic adaptation (Cherezhanova et al., 1971; Shevchenko et al., 1992), that is, chronic low-dose irradiation appears to alter the genetic structure of a population. Such a response to external damage, i.e., an expansion of the variability of the gene pool that provides a possible basis for the subsequent selection of the most adaptable forms, is a reflection of the fundamental mechanisms (providing the basis for life) (Geraskin, 1995) that ensure resistance of living systems and their possible adaptation to varying conditions of the environment.

5. Conclusions

Results of studies carried out in 1986–1989 on agricultural plants grown in the 10-km zone of the ChNPP revealed some fundamental features of the responses of the biota to severe radioactive accidents. In the most acute phase of exposure (spring–autumn of 1986 for the Chernobyl accident) the radiation damage to plant communities, as measured using basic cytogenetic tests, resembled the effect produced by acute γ-irradiation at similar doses. Since 1987, the incidence of cytogenetic disturbance has been elevated for several years, and this depended on the magnitude of the radioactive deposition despite the decrease in the radiation exposure with time. However, the rate of decline in cytogenetic damage to plants lagged considerably behind that of the radiation exposure. Chronic low dose exposure in regions affected by a serious radioactive accident can cause an inheritable destabilisation of genetic structures appearing, in particular, as an increase in cytogenetic damage and karyotypic variability in the offspring of irradiated organisms.

Acknowledgements

This work was supported by a contract between the European Economic Community and the International Union of Radioecology (No. ERBCIPDCT940802). The authors would like to thank Dr N.I.Sanzharova, Dr S.V.Krouglov, Dr A.S.Filipas, Mr B.G.Lisjanskij who contributed significantly to the development and maintenance of the field experiments in the 10-km zone of the ChNPP. We also acknowledge Dr D.Woodhead for discussion of experimental data and Dr B.Howard for critical reading of the manuscript.

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