ON THE EFFECTS OF LOW DOSES (0–1.2 Gy) BETA RADIATION ON ZEA MAYS SEEDS ON 12 DAYS PLANTLETS*

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Zea mays seeds with uniform genophond were irradiated with 90 Sr source in the 0–1.2 Gy range. We found that doses of β^- radiation lower than 0.6 Gy have a stimulating effect on the growth of the plantlets and that the doses between 0.6 and 1.2 Gy had an inhibitory effect. The β^- irradiation did not produce evident biochemical changes on early development stages.

Key words: low doses of β - radiation, Zea mays, plants growth, photoassimilatory pigments, nucleic acids.

1. INTRODUCTION

The biological effects of environmental stress and the physiological response of living organisms that allows their survival under stress conditions is a widely studied topic. Since the middle of the 20th century, ionizing radiations from radioactive isotopes have been investigated to determine their genotoxic impact on living organisms. Radionuclides are released into the environment from various sources such as planned discharges from the nuclear power industry, disposal of radioactive waste, medical use, nuclear weapons development and nuclear accidents. Ionizing radiations are able to cause toxically and geno-toxically effects on organisms, because radionuclides accumulate in biotic and abiotic components of the environment [1]. These can directly disturb metabolic processes, such as plant respiration, photosynthesis, growth, active transport as well as ionic balance and enzyme synthesis [2]. It is known that nuclear radiation can stimulate morphogenetic changes manifest in the early development stages [3]. Research studies revealed that the very low doses of ionizing radiations can stimulate cell proliferation [4-5] and this stimulating effect on cell proliferation affects the initial growth phase [6]. In order to progress in the evaluation of the environmental impact of ionizing

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radiation, it is necessary to establish the relationship between exposure (dose rate, accumulated dose) and the possible effects induced in living organisms (plants and animals). A low number of research studies are dedicated of biological effects induced by beta irradiation on living organisms.

In this study, we investigated the low doses of beta radiation influence on the *Zea mays* seeds and induced effects in early ontogenetic stages of plants obtained from germinated irradiated seeds.

2. MATERIALS AND METHODS

Young plantlets, obtained from the control and beta-irradiated seeds, were studied in laboratory experiments. The Zea mays seeds were irradiated one at a time in an irradiation chamber that was build for this purpose, with doses ranging between 0.12 and 1.2 Gy. The hole in the upper part fits a glass tube than can be easily inserted and extracted. The tube is used to place the seed in the proximity of the beta irradiation source. The schematic of the irradiation chamber is presented in Fig. 1. The dose debit through the glass tube, in the very location where the Zea mays seeds were placed one by one, was measured using a RFT – KD27012 dosimeter with an ion chamber. Batches of 30 seeds were irradiated for times ranging between 3 and 30 minutes with 3 minutes step.

The β^{-} source was ⁹⁰Sr and decays by the scheme:

$${}^{90}\mathrm{Sr} \xrightarrow{\beta^-} {}^{90}\mathrm{Y}, \quad T_{1/2} = 28.79\,\mathrm{y} \tag{1}$$

having $E_{\beta^-} = 546$ keV, with a branching ratio of 100% [7]. The daughter nucleus, ⁹⁰Y, is unstable as well. It decays by the scheme:

90
 Y $\xrightarrow{\beta^{-}}$ 90 Zr, $T_{1/2} = 64.00h$ (2)

with the energies, branching ratios and half-lives presented in Table1.



Fig. 1 – The beta-irradiation chamber.

The β	energies, branc	hing ratios and l	nalf-lives of the	⁹⁰ Y [7]
	$E_{\beta^{-}}$ [keV]	<i>I</i> _{β⁻} [%]	Half-life	
	93.83	0.0000014	64.00 h	
	519.39	0.0115	64.00 h	
	642.77	0.0018	3.19 h	
	2280.1	99 9885	64.00 h	

12 days old plantlets, obtained from the control and beta-irradiated seeds were studied. The irradiated seeds germinated on a support consisting of porous paper impregnated with deionized water, in darkness and closed Petri dishes. After germination the young plantlets growing was conducted in the same controlled conditions of temperature $(23 \div 24^{\circ}C)$, illumination (10 h: 14 h light/dark cycle) and the culture medium of young plantlets was 10 ml of deionized water daily.

After 12 days of plants growth spectrophotometrical measurements were done. The device we used was a CECIL 1000 spectrophotometer UV-VIS with quartz cuvetts. Biological material consisted of green tissue from seedling leaves, both for exposed samples and control. Assimilatory pigments level (chlorophyll **a**, chlorophyll **b** and total carotenoid pigments) in the *Zea mays* young plantlets were extracted in 85% acetone and calculated according to the Meyer Bertenrath's method modified by Ştirban [8]. The average nucleic acid level was measured using 6% perchloric acid extracts and the Spirin's method [9]. Plant individual length was measured with 1 mm precision. Statistical analysis was done on the plant lengths series.

3. RESULTS AND DISCUSSIONS

We noticed that toxicity symptoms led to brown spots covering the leaf surface for the highest beta irradiation dose used in this experiment, as presented in Fig. 2.

The lengths of the 12 days plantlets were carefully measured. The average lengths and the standard deviations were calculated for each batch of irradiated seeds. The confidence interval was calculated for each batch of plantlets using the Student test, for the confidence levels P = 90%, 95% and 99%. The results are presented in Table 2, together with the β^- irradiation doses.

Fig. 3 presents the average plant batch lengths for each irradiation dose. We found that small doses of β - radiation have a stimulating effect on the growth of the plantlets. The maximum stimulation was induced by 0.6 Gy. The average length of each batch was compared with the lengths of the control, using

Table 1



Fig. 2 - 12 days old Zea mays plantlets. Toxicity symptoms on leaf surface.

Statistical analysis on planticts length data											
Irradiation time [minutes]	0	3	6	9	12	15	18	21	24	27	30
Irradiation dose [Gy]	0	0.12	0.24	0.36	0.48	0.60	0.72	0.85	0.97	1.09	1.21
Average length [mm]	51.63	66.8	70.2	70.5	70.93	70.63	60.23	54	53.2	51.7	51.73
Standard deviation	19.20	22.06	19.33	23.26	20.38	23.90	20.63	16.73	14.46	18.62	20.69
Relative error	0.37	0.33	0.27	0.32	0.28	0.33	0.34	0.30	0.27	0.36	0.40
Confidence interval [mm] P = 90%	5.95	6.83	5.99	7.20	6.31	7.40	6.39	5.18	4.48	5.77	6.41
Confidence interval [mm] P = 95%	7.16	8.22	7.20	8.67	7.59	8.91	7.69	6.23	5.39	6.94	7.71
Confidence interval [mm] P = 99%	9.64	11.07	9.70	11.68	10.23	12.00	10.36	8.40	7.26	9.35	10.39

 Table 2

 Statistical analysis on plantlets length data

the Student test. The values of the equivalent dispersion and the t value of each pair are presented in Table 3.

We noticed that the average lengths of the batches irradiated with doses in the range 0–0.6 Gy are statistically significant bigger than the lengths of the control. Therefore we can conclude that the small doses of the β^- irradiation have a stimulating effect in respect to plant growing.



Fig. 3 - The average length versus low doses of beta irradiation.

Table 3

Comparison of average lengths of the batches irradiated with the control average length (* – statistically different, ** – statistically not different)

Irradiation dose [Gy]	0.12	0.24	0.36	0.48	0.60	0.72	0.85	0.97	1.09	1.21
S_e	20.68	19.27	21.33	19.80	21.68	19.93	18.01	17.00	18.91	19.96
t _{calc}	2.83	3.73	3.42	3.77	3.39	1.67	0.50	0.35	0.01	0.01
P = 90%, t = 1.676	*	*	*	*	*	**	**	**	**	**
P = 95%, t = 2.008	*	*	*	*	*	**	**	**	**	**
P = 99%, t = 2.677	*	*	*	*	*	**	**	**	**	**

The levels of the assimilatory pigments were comparatively studied on the basis of graphical representations of chlorophyll a, chlorophyll b and total carotenoids, Fig. 4 as well as by means of the chlorophylls ratio, Fig. 5. We found that the chlorophyll **a** to **b** ratio has a small variation with the irradiation dose. The total assimilatory pigments contents have the same variation with the beta-irradiation dose that was observed for all assimilatory pigments level. The chlorophylls ratio measurement offered the main insight into the photosynthesis complex processes since they revealed the response of the **LHC II** system (Light Harvesting Complex II) to the external stress.

We also found that increasing beta-irradiation dose does not induce considerable changes in chlorophylls ratio. The chlorophylls ratio values of the irradiated samples were within the error bars as compared with the control sample, suggesting that beta radiation does not significantly influence the photosynthesis process.



Fig. 4 – Assimilatory pigments level in Zea mays plantlets grown from low doses beta-irradiated seeds versus irradiation time.



Fig. 5 – Low doses of beta irradiation effect on the chlorophylls ratio in 12 days old *Zea mays* plantlets.

Another thing we found is a good linear correlation of all assimilatory pigments with the irradiation dose (two of these correlations are presented in Fig. 6). The average nucleic acid level of the irradiated samples decreases with the irradiation dose. Fig. 7 reveals this.

We found that beta irradiation does not produce evident biochemical changes on early development stages of *Zea mays* plantlets provided by irradiated seeds.



Fig. 7 – The average nucleic acid levels in Zea mays plantlets.

The basis of the biological effect of nuclear radiation is ionization, which can result in breakage of covalent bonds and damage of DNA, RNA, proteins and other molecules in the cell. The ions generated in this manner are reactive species that can react with biological molecules in the cell, causing damage. Due to their high propensity to react, the charged particles of beta radiation tend to lose their energy rapidly while passing through tissue, and are not highly penetrating. Diversity in dose-effect dependences due to low irradiation doses has been explained [10] as a change in the ratio between genetic damage and repair. According to Burlakova *et al.* [10], repairing systems are not activated by low doses, because it takes longer for them to get activated.

4. CONCLUSIONS

The low doses of beta irradiation revealed the stimulatory influence on the growth of plants, obtained from irradiated seeds, in their early ontogenetic stages.

Beta irradiation doses, ranging between 0.2 and 1.2 Gy do not produce significant biochemical changes in the early development stages of *Zea mays* plantlets obtained from irradiated seeds.

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