

**PHYSIOLOGICAL RESPONSE OF BEANS (*Phaseolus vulgaris* L.)
TO GAMMA-IRRADIATION TREATMENT.
II. WATER-EXCHANGE, RESPIRATION AND PEROXIDASE
ACTIVITY**

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Abstract. The aim of the current research was to study the tolerance of young bean plants (the cultivars Plovdiv 10 and Plovdiv 11) to gamma-irradiation stress with regard to water exchange, the respiration intensity and the peroxidase activity. For this purpose, the dry bean seeds were irradiated by Co⁶⁰ gamma-rays with doses of 150 and 200 Gy. On the 30th day of plant development, analyses of the water potential, the respiration intensity and the peroxidase activity were made. The results of the research show that the applied irradiation doses of 150 and 200 Gy decrease the water potential, and this is most evident in the cultivar Plovdiv 11. The applied doses of irradiation increase the respiration intensity and the peroxidase activity. There is an apparent cultivar response.

Keywords: cultivars beans, pre-sowingly irradiated seeds, water potential, respiration intensity, peroxidase activity.

AIMS AND BACKGROUND

In the physiological response of the plants to each stress influence, non-specific reactions, mobilizing the reserve capacities of the organism are being included. Some of them are connected with the respiration process and the water exchange. The respiration is the basic energy source, used both for synthetic processes and for the mobilization of compounds, non-specific to the plant organism. The respiration intensity is considered as a parameter determining the rate of plants growth^{1,2}. The existence of such a correlation indicates that the respiration is either a process, restricting the growth, or it undergoes changes parallel with this process.

In gamma-irradiation of wheat seeds and plants the respiration is being activated and along with this, some changes occur in positive aspect and in the peroxidase activity³.

In our previous research⁴ we found out that plants grown from irradiated seeds had inhibited root system and reduced transpiration. It was also found that under the conditions of disturbed water supply of the cells, there occurs accumulation of the amino acid proline⁵.

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The aim of the current study was to trace the changes in the respiration and the peroxidase activity in two bean cultivars of different origin – Plovdiv 10 and Plovdiv 11, treated pre-sowingly with gamma-rays Co^{60} – doses of 150 and 200 Gy. What is also of interest is the changes in the water exchange of the bean plants, and the content of free proline 30 days after the sowing of the irradiated seeds. The use of two genotypes had for an object not only to record the cultivars peculiarities, but mainly to enlarge the knowledge regarding the crop reaction to irradiation stress.

EXPERIMENTAL

Dry bean seeds, the cultivars Plovdiv 10 and Plovdiv 11, were irradiated with Co^{60} gamma-rays with doses of 150 and 200 Gy. The irradiated and control seeds were superficially treated with a 1% (w/v) solution of $\text{Ca}(\text{OCl})_2$ in a 10% (w/v) ethanol. The seeds germinated at a temperature of 22-24°C. They were sown in plastic vessels with a capacity of 0.5 l, there were four seeds in each vessel. Each variant consisted of five vessels. The plants were grown as substrate crops in a climatic box till they became 30-day old under the following conditions: light intensity 200 $\mu\text{mol m}^{-2}\text{s}^{-1}$ (PAR), photo period 14 h, temperature of 22±2/20±2°C day/night and relative humidity of the air 60-70%.

In the young, 30-day old bean plants of the two cultivars, the respiration intensity and the peroxidase activity after Boyarkin⁶ were determined, the water potential of the leaves was measured using a pressure technique chamber⁷. The free proline content was determined in youngest fully leaves⁸. Relative water content (RWC) was determined in % (Ref. 6).

RESULTS AND DISCUSSION

Tables 1 and 2 show the results of the gamma-irradiation influence on the water status of the bean plants. It is evident that in plants of the cultivar Plovdiv 10 RWC decreases by 7% at irradiation of the seeds with 150 Gy and by 13% with 200 Gy. Almost the identical tendency is observed in the second cultivar – Plovdiv 11 – by 6 and 14% for the two irradiation doses, respectively.

The plants water potential (Ψ), which is the basic thermodynamic value of the water exchange, decreases considerably – by 31 and 45% at irradiation doses of 150 Gy for the cultivars Plovdiv 10 and Plovdiv 11, respectively. In plants, grown from seeds treated pre-sowingly with 200 Gy, these values are considerably lower than the control – 92% in the cultivar Plovdiv 10 and 120% in the cultivar Plovdiv 11.

The changes in RWC and water potential probably are a result of the disturbances in the water exchange and the plants adaptation caused by the irradiation stress.

Table 1. Influence of gamma-irradiation on the water status in bean plants – cultivar Plovdiv 10

Indices	0 Gy	150 Gy	200 Gy
PWC	85.96±1.6	79.94±1.9* (93)	75.33±1.6*** (87)
Ψ	-0.29±0.02	-0.39±0.01*** (131)	-0.56±0.02*** (192)
P	5.68±0.21	12.86±0.82** (226)	21.40±1.12*** (376)

* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; in brackets – % difference from the control; PWC – relative water content (%); Ψ – water potential (MPa); P – proline content in leaves (mg g^{-1} free tissue).

It is a well-known fact that in case of water deficiency in the plant cells there occurs a process of osmotic self-regulation directed toward a preservation of the water balance. Under these conditions free amino acids are being accumulated – mainly proline, which may increase its amount dozens of times.

Table 2. Influence of gamma-irradiation on the water status in bean plants – cultivar Plovdiv 11

Indices	0 Gy	150 Gy	200 Gy
PWC	84.36±2.1	79.22±0.95* (94)	73.11±1.4*** (86)
Ψ	-0.26±0.02	-0.38±0.01*** (145)	-0.58±0.03*** (220)
P	5.90±0.42	13.50±1.06** (228)	24.60±1.15*** (416)

* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; in brackets – % difference from the control; PWC – relative water content (%); Ψ – water potential (MPa); P – proline content in leaves (mg g^{-1} free tissue).

At the irradiation dose of 150 Gy the increase of the free proline content in plants of the two cultivars shows a similar tendency – the content increases twice. More considerable differences are observed at the higher dose of 200 Gy. In the plants of the cultivar Plovdiv 10 the free proline content increases 3.7 times in comparison with the control, and in plants of the second cultivar this correlation is 4.6. The data showing the gamma-rays influence on the respiration intensity and the peroxidase activity in young bean plants – the cultivars Plovdiv 10 and Plovdiv 11 – are presented in Tables 3 and 4.

The results in Table 3 and 4 indicate that the applied two irradiation doses (150 and 200 Gy) increase the respiration intensity in the leaves by 12 and 41% for the cultivar Plovdiv 10 and by 18 and 38% for the cultivar Plovdiv 11, respectively. This is probably due to the energy deficiency which occurred as a result of the inhibited photosynthesis. The supposition is confirmed by the data on the peroxidase activity.

Table 3. Influence of gamma-irradiation on the respiration intensity R ($\text{mg CO}_2\text{g}^{-1}\text{ fr. wh}^{-1}$) and the peroxidase activity (PA – U – g fr. w min^{-1}) in bean plants of the cultivar Plovdiv 10

Indices	0 Gy	150 Gy	200 Gy
R	0.625±0.021	0.698±0.03* (112)	0.883±0.016** (141)
PA	826±32	963±16** (116)	1320±41*** (159)

* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; in brackets – % difference from the control.

In plant organisms, subjected to stress, the activity of the antioxidising enzymes increases. Peroxidase is one of these enzymes. It was found that apart from the cell walls and the cytoplasm, this enzyme is also located in the mitochondrias and the chloroplasts, i.e. where the irradiation has the strongest influence⁹. The peroxidase functional activity is connected with the neutralization of the harmful oxygen radicals which number is increased under stress conditions, particularly in the case of gamma-irradiation stress.

Table 4. Influence of gamma-irradiation on the respiration intensity R (mg CO₂g⁻¹fr. w h⁻¹) and the peroxidase activity (PA – U – g fr. w min⁻¹) in bean plants of the cultivar Plovdiv 11

Indices	0 Gy	150 Gy	200 Gy
R	0.610±0.015	0.720±0.012** (118)	0.840±0.072*** (138)
PA	810±26	986±22*** (122)	1410±52*** (174)

* $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; in brackets – % difference from the control.

The results indicate that in the case of moderate stress (150 Gy), the peroxidase is activated by 16% for the cultivar Plovdiv 10 and 22% for the cultivar Plovdiv 11, and in the case of a strong stress (200 Gy) with 50 and 74% above the control, respectively. This fact confirms the studies made by Khanna³. When comparing the data from the Tables above, we may sum up that with regard to these indices, the cultivar Plovdiv 10 is more tolerant to gamma-irradiation stress than the cultivar Plovdiv 11, which is more radio-sensitive.

CONCLUSIONS

Gamma-rays, applied pre-sowingly to bean seeds exert influence on the water exchange parameters, the influence being almost identical for plants of the two genotypes. RWC is decreased to a slighter degree, while Ψ goes down, more strongly for the cultivar Plovdiv 11.

More considerable differences are recorded with regard to the respiration and the peroxidase activity. For the two indices, the differences from the control are greater for the plants from the cultivar Plovdiv 11.

As a result of the gamma-irradiation, the proline content in the plant cells is considerably increased. Cultivar sensitivity occurs.

Cultivar Plovdiv 10 is more tolerant to gamma-irradiation stress than the cultivar Plovdiv 11, which is more radio-sensitive.

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