

## **GAMMA RADIATION EFFECT ON SUPEROXIDE DISMUTASE ACTIVITY IN *HYPERICUM PERFORATUM* L. AND *ECHINACEA PURPUREA* L., MOENCH.**

**VLAD ARTENIE<sup>1\*</sup>, DANIELA LUMINIȚA ICHIM<sup>1\*</sup>,  
GOGU GHIORGHITA<sup>2</sup>, MIHAELA PINTILEI<sup>1</sup>**

**Keywords:** gamma radiation, superoxide dismutase, *Echinacea purpurea* L., Moench, *Hypericum perforatum* L.

**Abstract :** In this paper we were focused on the activity of superoxide dismutase (SOD) in *Hypericum perforatum* L and *Echinacea purpurea* L., Moench plantlets, obtained from seeds irradiated with gamma rays before germination. Total activity as well as specific activity of SOD in *Hypericum perforatum* L. plantlets shows an inhibition, which becomes higher at higher irradiation doses. For *Echinacea purpurea* L. species, gamma radiation shows a slowly stimulative effect of total activity of SOD for some of the higher doses, but the specific enzyme activity is inhibited for all irradiation doses applied on non-germinated irradiated seeds.

### **INTRODUCTION**

Ionizing radiation has a multiple and various effect on the living organisms. They can stimulate or inhibit the physiological and biochemical processes, which leads to a deficiency which can affect the normal cell growth and its normal evolution. [6]. The action of the ionizing radiation in the living cells is associated with formation and accumulation of toxic combinations, such as the reactive species of the oxygen [4]. This association represents a potential danger for cellular integrity, being capable to cause the malfunction of the metabolic processes, inclusive the modification of enzymatic activity which is involved in this process [7]. In the living cell there are specific biochemical mechanisms of protection which permit transformation of the reactive species of the oxygen in non-toxic complexes [2, 3]. For example, superoxide dismutase convert superoxide in hydrogen peroxide and catalase transforms hydrogen peroxide in water and molecular oxygen.

In this work we study gamma radiation effect on superoxide dismutase activity (EC 1.11.1.5) from plantlets of two medicinal plants, *Hypericum perforatum* L. and *Echinacea purpurea* L., Moench., plantlets which comes from seeds which were irradiated before germination.

### **MATERIALS AND METHODS**

Biological material was represented by 14 days old plantlets from seeds of *Hypericum perforatum* L and *Echinacea purpurea* L., Moench. The seeds of *Hypericum perforatum* L. were provided the Secuieni Agro-Zootechnical Research Station, Neamț Country and those of *Echinacea purpurea* L., Moench by the Research and Development Centre for Potatoes and Sugar Beets from Brasov, 2003 harvested. The seeds were irradiated in the Nuclear Department of Chemical-Pharmaceutical Research Institute (ICCF) Bucharest. The irradiation ratio was 5Gy/minute and the irradiation doses were: 10Gy, 30Gy, 50Gy, 75Gy, 100Gy, 120Gy, 150Gy for *Hypericum perforatum* L and 10Gy, 30Gy, 50Gy, 75Gy, 100Gy, 120Gy for *Echinacea purpurea* L., Moench.

The germination was performed in the laboratory conditions. Activity of superoxide dismutase was determined using the spectrophotometric method [7], with the help of the potential enzyme to inhibit the decrease of nitro-blue-tetrazolium by the superoxide radicals resulted in the medium of reaction by riboflavin photoreduction. One unit of superoxide dismutase represents the quantity of enzyme which produces 50% inhibition in the standard conditions. Specific activity it is shown in enzymatic units per milligram of protein.

The dosage of soluble proteins in enzymatic extracts was realized by Bradford method [1], using bovine serum albumin as a standard.

The experimental data was statistically analyzed using the Student test [9].

### **RESULTS AND DISSCUSIONS**

For *Hypericum perforatum* L. and *Echinacea purpurea* L. plantlets obtained from seeds which were irradiated before germination with gamma radiations, our research shows that

activity of superoxide dismutase (SOD) fluctuates according with the irradiation dose which was used for each sample.

Figure 1 shows the activity of superoxide dismutase in the *Hypericum perforatum* L. plantlets, plantlets which comes from pre-irradiated seeds with doses between 10 and 150 Gy. The activity of superoxide dismutase varies between 4994,235 – 7096,722 enzymatic units(UE) / g fresh material in comparison with comparative 7141,083 UE/g values which represent non-irradiated control.

Related to the control it is easily obvious that SOD activity decreases on the irradiated samples, fact which concludes that irradiation had an inhibitory effect on the enzyme.

SOD activity records a minimum value to the 30Gy dose, being equivalent with only 70% of control enzyme activity; this difference is very strongly statistical represented.

A significant decrease of the SOD activity is obtained for 50 and 150 Gy doses.

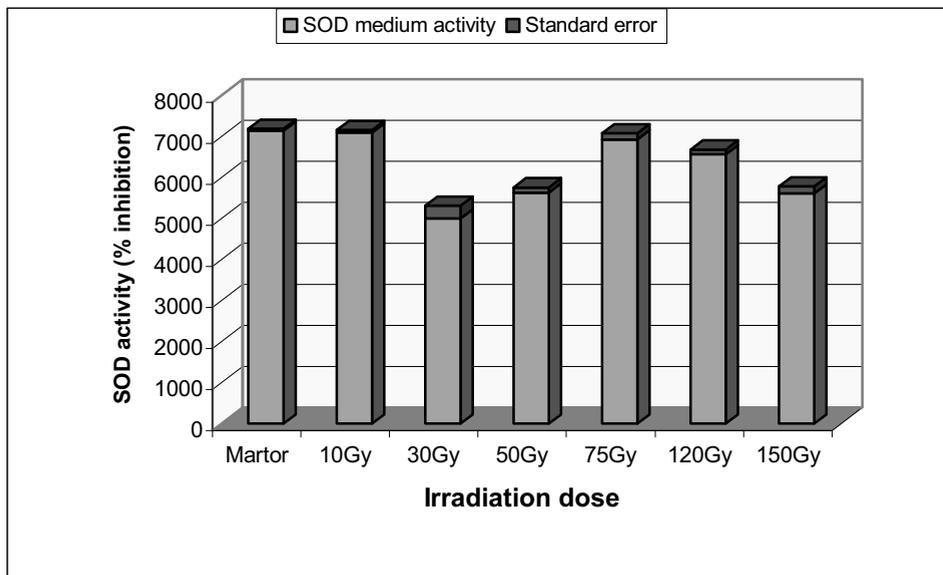


Figure 1: Superoxide dismutase activity (% inhibition/g) of *Hypericum perforatum* L. plantlets obtained from irradiated seeds.

Net inhibitory effect of gamma radiation on SOD activity from *Hypericum perforatum* L. plantlets it is confirmed from the evolution of specific activity (UE/mg protein). This specific activity was obtained by reporting the total activity to the quantity of water soluble proteins from the enzymatic extract. (Figure 2).

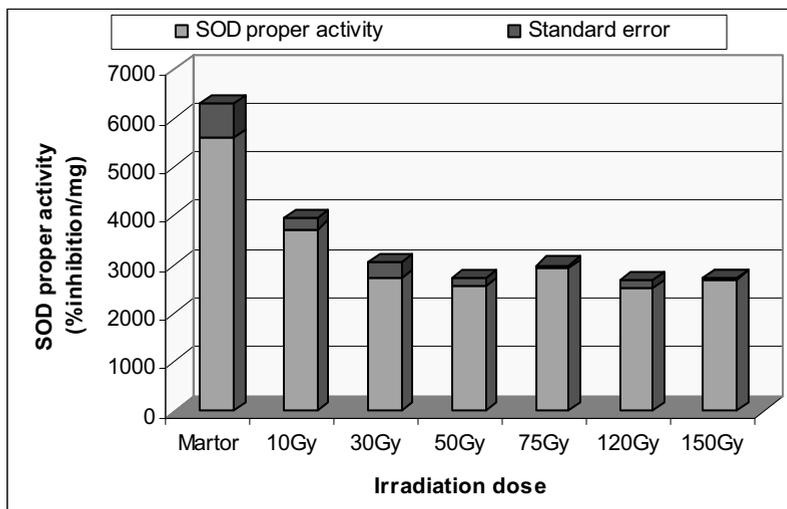


Figure 2: Superoxide dismutase specific activity (% inhibition/mg of protein) in plantlets of *Hypericum perforatum* L. obtained from irradiated seeds

As we can notice from the Figure 2, SOD specific activity in *Hypericum perforatum* L. plantlets decrease to 67% from the control value, for 10 Gy dose, and for the rest of the doses to a value between 46-52% compared with non-irradiated plantlets.

Superoxide dismutase activity for *Echinacea purpurea* L., Moench. plantlets recorded values between 6243-7770 UE/g (%inhibition/g).

By comparison with control activity of 6358 UE/g, irradiation of seeds leads to a small inhibition of the enzyme : only 9% for 75Gy dose and 22% for 100Gy dose (Figure 3). The rest of dose treatment was associated with an inhibition of SOD activity.

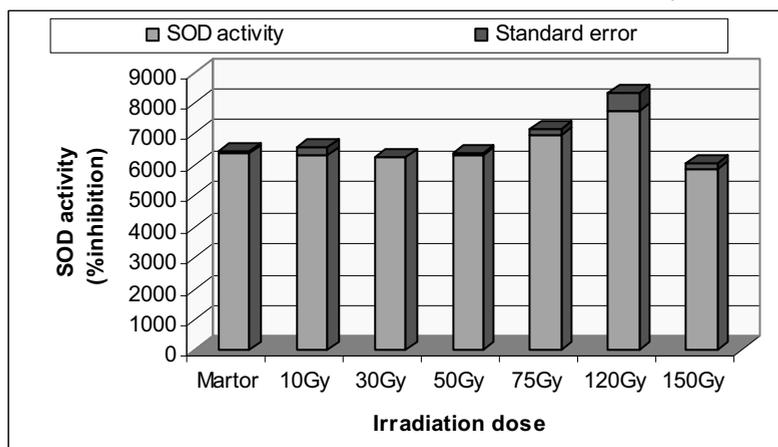


Figure 3: Superoxide dismutase activity (% inhibition/g) for *Echinacea purpurea* L., Moench plantlets which came from irradiated seeds.

Dosing of water soluble proteins from enzymatic extract, and determine the values for specific activities of enzyme (% inhibition/mg proteins) reveals that the enzyme activity is inhibited for all irradiation doses (Figure 4).

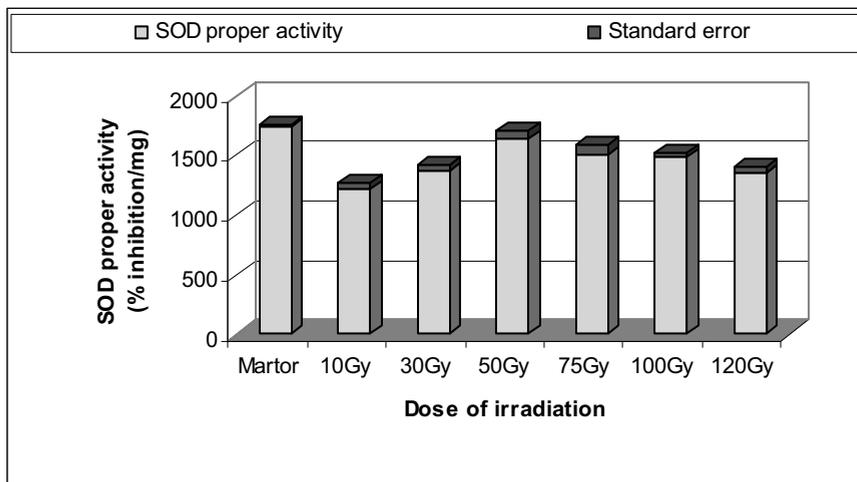


Figure 4: Superoxide dismutase specific activity (% inhibition/mg protein) for *Echinacea purpurea* plantlets, which came from irradiated seeds.

This inhibition at lower doses has a more prominent effect than those at higher doses. For 50 Gy dose, specific activity of SOD shows a non-significant decrease compared with control value (Figure 4).

Our results are similar with those already published in related papers [5, 6, 8], in the sense that the high doses of irradiation lead to an inhibiting of enzymatic activity and, those low ones could lead to slight stimulation but in certain conditions lead to a slight inhibiting of enzymatic activity. Up to now, based on different papers [5,6,8], studies of modification of the enzymatic activity by irradiation have contradictory results. In seeds of *Hypericum perforatum* L. and *Echinacea purpurea* (L.) Moench, which were irradiated pre-germination with gamma radiation, peroxidases and catalase activity varies differently according to species and to irradiation dose [2]. Gamma radiations have a significant effect on the catalase activity also in the young plants of *Quercus robur*, the enzyme response being direct proportional with plant age and irradiation time [3]. Ghiorghita[5] found that the increase in the activity of catalase and peroxidases both to low and high doses in their studies on wheat (*Triticum aestivum*) and two-row barley (*Hordeum distichum*).

## CONCLUSIONS

SOD activity for *Hypericum perforatum* L. plantlets, obtained from seeds which were gamma irradiated pre-germination, suffers a stronger inhibition for the higher dose of radiations.

Also, gamma radiations has an inhibitory effect, effect which became stronger for 50 and 75 Gy doses on plantlets of *Echinacea purpurea* L., plantlets which came from pre-germination irradiated seeds.

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<sup>1</sup> „Alexandru Ioan Cuza” University, Iasi, Biology Faculty, Biochemistry Department.

<sup>2</sup> „George Bacovia” University, Bacau, Science Faculty, Genetics Department.

\* - vartenie@uaic.ro

