

THE INFLUENCE OF CERTAIN TREATMENTS WITH PHYSICAL MUTAGENE AGENTS ON THE ACTIVITY OF SOME ENZYMES IN *ECHINACEA PURPUREA* AND *HYPERICUM PERFORATUM*

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Abstract: We determined catalase and peroxidase activity in *Echinacea purpurea* and *Hypericum perforatum* plantlets resulted after the germination of seeds irradiated with radiation doses of 1, 3, 5, 8, 10, 12, 15 and 20 KRad, the radiation flow being of 0.5 KRad per minute. After interpreting the results obtained we found the radiation influence on enzyme activity, in the respect of its stimulation at low radiation doses.

INTRODUCTION

The catalase (E.C. 1.11.1.6) and peroxidase (E.C. 1.11.1.7) are di-component enzymes that belong to the haemoprotein class, having the heme as prosthetic group, that is the IX ferri-porphyrine.(Artenie and Tanase,1981).

These oxido-reducing enzymes have a significant biological role, being involved in the protection against stressful factors, such as radiation, pesticides a.s.o.(Corneanu,1989). In the specialised literature there are lots of data on the influence of radiation over vegetal organisms, in the respect of causing oxidative stress, as suite of formation of reactive oxygen species.(Cojocaru,1997)

Purpose of the research

This works aims to obtain certain data regarding the dynamics of certain enzymes' activity in the *Echinacea purpurea* and *Hypericum perforatum* plantlets,after their treatment with gamma radiation at different radiation doses.

This is furthermore justified by the well-known fact that these herbs are extremely appreciated for their therapeutic qualities, among which the antiviral effect shows up by itself, envisaging the acquire of immune-stimulating, anti-inflammatory, scarring and anti-HIV drugs.(Ciulei et al,1993).

MATERIAL AND METHODS

The research was carried out on 14-days plantlets obtained from irradiated seeds that were put to germinate in controlled conditions.

The biological samples come from the Secuieni Agro-Zootechnical Research Facility, Neamț County, and the seeds were irradiated at the Nuclear Unity of ICCF Bucharest with increased radiation doses, at the flow of 0.5 KRad/min.

The catalase activity has been determined by iodine titration.(Artenie and Tanase,1981) and the peroxidase activity by the colorimetric method with o-dianisidine(Möler and Ottolenghi ,1966).

The extraction of catalase from the biological material was made by Na₂HPO₄ solution 0.1M, and that of the peroxidase with phosphate buffer 0.4M.

We carried out 4 to 6 determinations for each sample and witness separately, and the results we obtained were statistically processed by using the STUDENT test.

RESULTS AND DISCUSSIONS

The experiments performed have outlined the influence of radiation on the enzyme activity in the analysed samples.

Table I: Catalase activity(CU/g/min) in *Echinacea purpurea*:

Sample	N	Average	Standard error	Probability
M	4	3.5646	±0.1948	-
P1	4	3.6120	±0.0490	p>0.5
P3	4	4.0156	±0.1063	0.5>p>0.25
P5	4	5.8100	±0.1722	0.01>p>0.002
P8	4	5.4750	±0.0333	0.002>p>0.001
P10	4	2.9746	±0.1917	0.5>p>0.25
P15	4	6.4600	±0.1294	0.01>p>0.002
P20	4	6.2046	±0.0649	0.002>p>0.001

Legend: M = non-irradiated witness
 P1 = sample with 1 KRad radiation dose
 P3 = sample with 3 KRad radiation dose
 P5 = sample with 5 KRad radiation dose
 P8 = sample with 8 KRad radiation dose
 P10 = sample with 10 KRad radiation dose
 P15 = sample with 15 KRad radiation dose
 P20 = sample with 20 KRad radiation dose
 n =no. of assessments

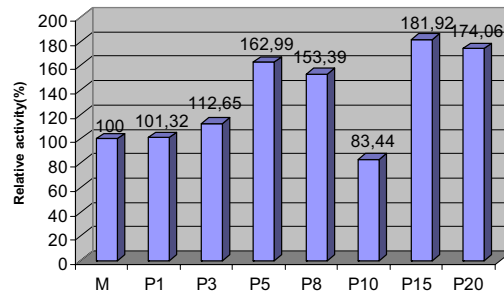


Fig.I: Relative activity(%) of catalase in *Echinacea purpurea*

Legend: idem Table I

As one can see, at low radiation doses (1, 3 KRad respectively), the activity is similar to that of the witness, while in the P5 and P8 samples one can see a strong stimulation of the enzyme activity.

Table II: Peroxidase activity(PU/g/min) in *Echinacea purpurea*:

Sample	n	Average	Standard error	Probability
M	4	2.0058	±0.0744	-
P1	4	2.6398	±0.0856	P<0.001
P3	4	2.9656	±0.0919	P<0.001
P5	4	2.5346	±0.0848	0.5>P>0.25
P8	4	2.1358	±0.1356	0.01<P<0.002
P10	4	2.0156	±0.0185	0.05>P>0.02
P15	4	1.9658	±0.0068	0.25>P>0.1
P20	4	2.0282	±0.1256	0.01>P>0.002

Legend: M = non-irradiated witness
 P1 = sample with 1 KRad radiation dose
 P3 = sample with 3 KRad radiation dose
 P5 = sample with 5 KRad radiation dose
 P8 = sample with 8 KRad radiation dose
 P10 = sample with 10 KRad radiation dose
 P15 = sample with 15 KRad radiation dose
 P20 = sample with 20 KRad radiation dose
 n =no.of assessments.

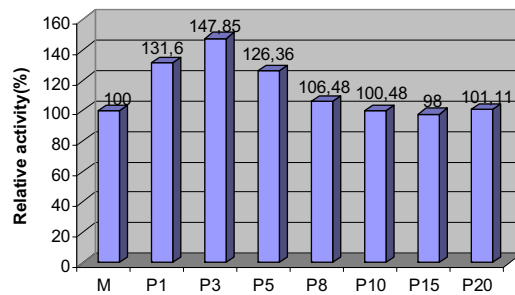


Fig. II:Relative activity(%)of peroxidase in *Echinacea purpurea*

Legend: idem Table II.

The peroxidase activity for the species of *Echinacea purpurea* recorded a significant increase at low radiation doses (1 and 3 KRad), opposite to the catalase, while at high doses the activity becomes similar to that of the witness.

Table III:Catalase activity(CU/g/min) in *Hypericum perforatum*:

Sample	n	Average	Standard error	Probability
M	4	0.7556	±0.0929	-
P1	4	1.3173	±0.0185	0.05>p>0.02
P3	4	1.0270	±0.0069	0.25>p>0.1
P5	4	0.9663	±0.0251	0.5>p>0.25
P8	4	0.9423	±0.0241	0.5>p>0.25
P12	4	1.2446	±0.0184	0.05>p>0.02
P15	4	0.3223	±0.0121	0.1>p>0.05

Legend: M = non-irradiated witness
 P1 = sample with 1 KRad radiation dose
 P3 = sample with 3 KRad radiation dose
 P5 = sample with 5 KRad radiation dose
 P8 = sample with 8 KRad radiation dose
 P12 = sample with 12 KRad radiation dose
 P15 = sample with 15 KRad radiation dose
 n =no.of assessments

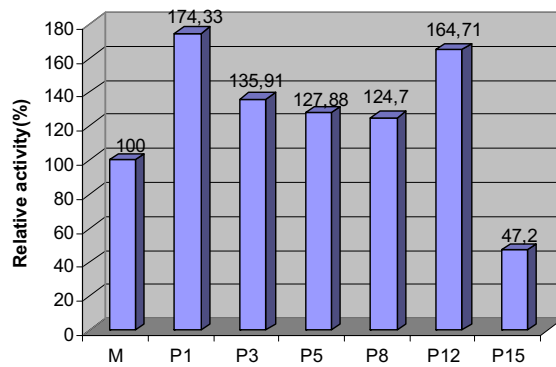


Fig. III:Relative activity(%)of catalase in *Hypericum perforatum*

Legend: idem Table III

From the above data we find the catalase activity was strongly stimulated by small radiation doses, whilst at 15 KRad the enzyme's activity was strongly inhibited, thus recording a much lesser value compared to the witness.

Table IV: Peroxidase activity(PU/g/min) in *Hypericum perforatum*:

Sample	n	Average	Standard error	Probability
M	6	4.2085	±0.0944	-
P1	6	6.5912	±0.1022	p<0.001
P3	6	7.6755	±0.2651	p<0.001
P5	6	3.0757	±0.0848	0.01<p<0.002
P8	6	4.4526	±0.1383	p<0.5
P12	6	4.1466	±0.0937	p>0.5
P15	6	6.1818	±0.2226	0.05<p<0.02

Legend: M = non-irradiated witness
 P1 = sample with 1 KRad radiation dose
 P3 = sample with 3 KRad radiation dose
 P5 = sample with 5 KRad radiation dose
 P8 = sample with 8 KRad radiation dose
 P12 = sample with 12 KRad radiation dose
 P15 = sample with 15 KRad radiation dose
 n =no.of assessments

Subsequent to the research undertaken on *Hypericum perforatum*, we found that peroxidase activity was strongly stimulated at doses of 1, 3 KRad respectively, inhibited at 5 KRad, whilst high doses of 15 KRad caused a new increase in enzyme's activity.

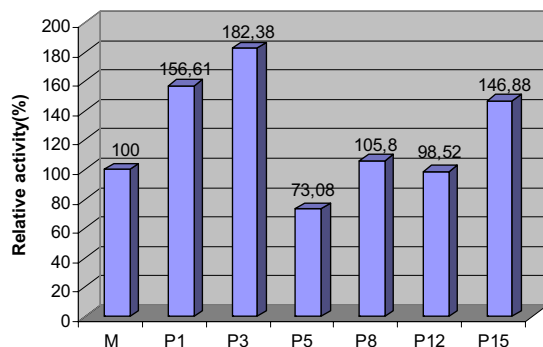


Fig. IV: Relative activity(%)of peroxidase in *Hypericum perforatum*

Legend: idem Table IV.

CONCLUSIONS

Low radiation doses caused an important increase in peroxidase activity in *Echinacea purpurea*, while the catalase activity in the same species recorded a significant increase at doses of 5, 8, 15 and 20 KRad respectively.

In the species *Hypericum perforatum*, we found a stimulation of peroxidase activity at 1, 3 KRad, while catalase activity is increased at low radiation doses and strongly inhibited at 15 KRad.

The results obtained are similar to those in the specialized literature, low radiation doses causing a stimulation of enzyme activity.

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