THE SEASONAL DYNAMICS OF CATALASE AND PEROXIDASE IN EVERGREEN PLANTS

VLAD ARTENIE[1*], CIPRIAN DARABAN[1], MIHAELA IFTINCA[1]

Keywords: catalase, peroxidase, seasonal rhythm, *Picea pungens*, *Thuja occidentalis*

Abstract: The study analyzes the results recorded on the dynamics of catalase and peroxidase activity in the leaves of the evergreen plants *Picea pungens*, variety argentaea, and *Thuja occidentalis*, variety fastigata, during the four seasons of a calendar year. The catalase activity in the leaves of *Picea pungens* is maximum during summer, while that observed in the *Thuja occidentalis* leaves is higher in the autumn and first winter months. Peroxidase activity in the leaves of the two species following quite a similar pattern. The *Picea pungens* leaves show a less active peroxidase in winter and respectively, springtime while, in the case of *Thuja occidentalis*, its activity is oscillating, the maximum being recorded in the beginning of August. Further on, peroxidase activity gets reduced, so that the values recorded in October and November are quite close to those of the spring months.

INTRODUCTION

Catalase (EC 1.11.1.6) and peroxidase (EC 1.11.1.7) play a special role in the substance metabolism plants, as these oxidoreductases are involved in the decomposition of hydrogen peroxide (H_2O_2) that may occur in the vegetal cell under the action of different stress factors. The two oxidoreductases represent bicomponent enzymes, belonging to complex proteins. To the chemical structure of catalase and peroxidase there belongs – as a prosthetic group – the iron (III) – protoporphyrine IX (hemine). Such a combination – which is structurally related to chlorophyll – is synthesized in the living organisms by the same precursors, on a common path, the stage of protoporphyrine IX's formation included.

Previous investigations of ours [Artenie et al., 1998] have demonstrated that the level of chlorophyllian pigments in the leaves of *Picea pungens* and *Thuja occidentalis* plants differs from one season to another.

In the present study, the seasonal dynamics of catalase and peroxidase activity in *Picea pungens* and *Thuja occidentalis* leaves will be followed along the four seasons of a calendar year.

MATERIAL AND METHODS

The dynamics of catalase and peroxidase activity has been studied in the leaves of the *Picea pungens*, variety argentaea and, respectively, *Thuja occidentalis*, variety fastigata, growing in the "Anastasie Fătu " Botanical Garden of the " Alexandru Ioan Cuza" University of Iaşi, between December 1, 2001 – November 30, 2002.

The leaves considered for investigating the two enzymes activity have been harvested in the last two days or first three days of each month, at the same morning hours (between 7.30 and 8.00), with the exception of the month of July, when they have been taking over on the day of 26. From the freshly – collected leaves, enzymatic extracts have been prepared in the laboratory, under determined conditions.

The catalase activity has been determined by the titrimetric method, with sodium thiosulfate [Artenie, Tănase, 1981], while peroxidase activity has been estimated by the photocolorimetric method, on using orto – dianisidine as a hydrogen donor [Möller and Ottolenghi, 1966]. In all cases, for determining the activity enzymes, five leaf samples have been taken over from each of the species under study.

The experimental data obtained have been processed statistically on the basis of Student's test [Snedecor, 1968]. Data are presented as the means ±SE. Differences were considered significant at $p < 0.05$.

RESULTS AND DISCUSSIONS

The investigations performed evidenced that, in the leaves of *Picea pungens*, variety argentaea, and respectively, *Thuja occidentalis*, variety fastigata, the activity of catalase and peroxidase records fluctuant values along the four seasons of a year.

As seen from the Fig.1, catalase activity in the *Picea pungens* leaves increases progressively until midsummer, when the maximum level is recorded by the end of July. In August, it remains high and it is only by the end of September that the catalase activity returns to a value comparable to that recorded in springtime. In the months to follow, it gradually decreases, attaining approximately the same specific stage in December 2001 and 2002.

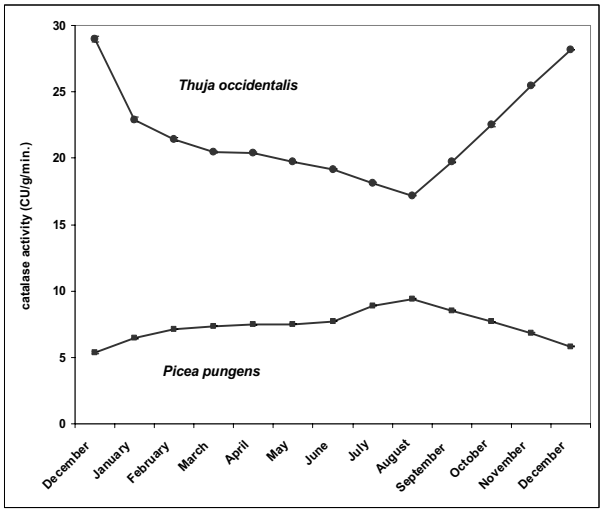

Fig.1. Annual dynamics of catalase activity (CU/g/min.) in the leaves of *Picea pungens*, variety argentaea (■—■) and *Thuja occidentalis*, variety fastigata (▲—▲) ; dates of samples prelevation : December 1, 2001; December 30, 2001; January 31, 2002 ; March 2, 2002 ; April 1, 2002 ; April 30, 2002 ; May 31, 2002 ; June 3, 2002 ; July 26, 2002 ; September 2, 2002 ; September 30, 2002 ; November 1, 2002 ; November 30, 2002.

According to some authors [Hlebnikov et al., 1963], the catalase activity in conifer species is lower and more stable during winter, comparatively with other seasons of the year. Beginning with the springtime, the activity of catalase increases, the maximum being reached in the second half of the summer, while, with the advance of autumn, the enzyme becomes less active [Hlebnikov et al.,1963].

As to the evolution of catalase activity in the *Thuja occidentalis* leaves, the observation to be made for this specie is that the enzyme is more active in the autumn and first winter months. Thus, catalase activity is higher in December 2001, followed by a gradual, slight decrease, the minimum value being recorded in the end of July. Further

on, the activity of catalase in the *Thuja occidentalis* leaves increases from one month to another, attaining, on December 1, 2002, the value recorded for December 1, 2001.

A comparison between the activities of catalase for the two species under study evidences that the values obtained are asymmetrical, with an opposite dynamics, which means that the maximum and minimum values are reversed.

As to the annual dynamics of peroxidase activity in the *Picea pungens* and *Thuja occidentalis* leaves, worth mentioning are the following observations. With both species, the peroxidase activity evolves according to a similar pattern (Fig. 2). The leaves of *Picea pungens* have a less active peroxidase during winter and, respectively, spring. With the installation of summer, the peroxidase activity in the leaves of *Picea pungens* gradually increases, the maximum value being attained in July-August. During autumn months, peroxidase remains sufficiently active until the end of November 2002. Interestingly enough, the activity of peroxidase recorded in December 2002 is about one unit higher than that of December 2001. If considering the curve profile, one may assert that the minimum activity of peroxidase in Picea pungens is recorded in winter.

In the *Thuja occidentalis* specie, the peroxidase activity shows a more oscillating evolution, comparatively with the preceding one. Thus, the peroxidase activity increases suddenly in the beginning of spring, quite close values being recorded for the months of March, April and May. Advancing towards to aestival season is characterized by a stimulation of peroxidase in the *Thuja occidentalis* leaves, the enzyme activity being maximum in the beginning of August. Further on, this is seen as decreasing, the values recorded in October and November being quite close to those of the spring months.

On December 1, 2002, the peroxidase evidences only half of the activity recorded in the autumn months, the levels recorded in December – January representing the minimum of peroxidase activity recorded in *Thuja occidentalis*. Once again, mention should be made of the fact that, on December 1, 2002, the peroxidase activity is double, comparatively with the values recorded exactly one year ago (December 1, 2001). Such a lack of agreement, evidenced for both species, is quite difficult to explain. Probably, the different temperatures (- 0.3°C and, respectively, - 8.5°C), as well as the amount of precipitation recorded in the months of December of both years (of 77.9 % and, respectively, 60.5 %) might provide an explanation.

The higher values of peroxidase activity registered in the leaves of the two species on November 30, 2002, comparatively with the level of enzyme activity on December 1, 2001, might be possibly explained by lowering of air temperature from – 0.3°C to – 8.5°C in the end of the period of samples taking over ; such an increase in peroxidase activity might be interpreted as a defense reaction of plants against low temperatures [Kawashima, Uritani, 1963 ; Rubin, Loginova, 1965].

The data collected in the study evidence that the *Thuja occidentalis* leaves posses a more active catalase than the *Picea pungens* ones. Peroxidase activity is more pronounced in the leaves of the *Picea pungens* species, comparatively with that of the enzyme occurring in the *Thuja occidentalis* leaves.

The different behavior of the catalase and, respectively, peroxidase, recorded in *Picea pungens* and *Thuja occidentalis* along the four seasons of the year may be obviously

explained through the genetic peculiarities of the two species, seen as differing between them by the genome each one possesses.

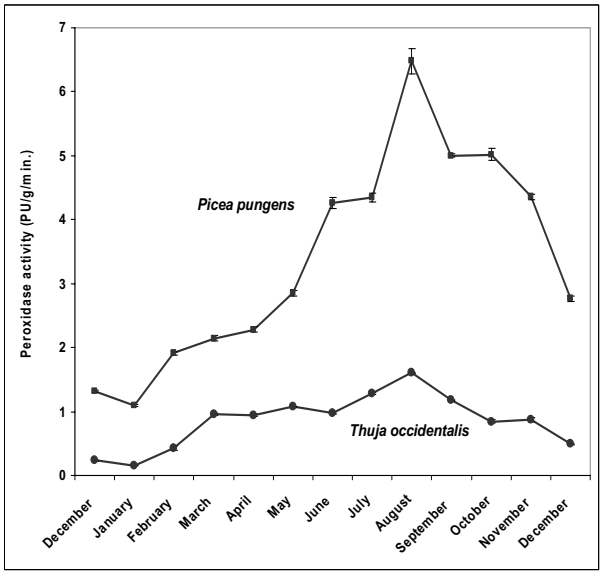

Fig. 2. Annual dynamics of peroxidase activity (PU/g/min.) in the leaves of *Picea pungens*, variety argentaea (■—■) and *Thuja occidentalis*, variety fastigata (▲—▲); dates of samples prelevation : December 1, 2001; December 30, 2001; January 31, 2002; March 2, 2002 ; April 1, 2002 ; April 30, 2002 ; May 31, 2002 ; June 3, 2002 ; July 26, 2002 ; September 2, 2002 ; September 30, 2002 ; November 1, 2002 ; November 30, 2002.

CONCLUSIONS

The catalase activity in the *Picea pungens* leaves increases progressively from December until midsummer, when the maximum level is recorded by the end of July.

In the *Thuja occidentalis* leaves, the catalase activity is more active in the autumn and first winter months. The minimum value of catalase activity is recorded in the end of July.

The leaves of *Picea pungens* have a less active peroxidase during winter and, respectively, spring. With the installation of summer, the peroxidase activity in the leaves of *Picea pungens* gradually increases, the maximum value being attained in July-August.

In the *Thuja occidentalis* leaves, the peroxidase activity is maximum in July-August. The levels in December – January represent the minimum of peroxidase activity recorded in *Thuja occidentalis*.

BIBLIOGRAPHY

Artenie Vlad, Tănase Elvira, 1981 – Practicum de biochimie generală, Centrul de Multiplicare al Universităţii „ Alexandru Ioan Cuza ” din Iaşi, p.135

Artenie V., Godja Anca, Ungureanu E., Ciurcă Mihaela, 1999 – The Seasonal Dynamics of the Photosynthetic Pigments in Several Evergreen Plants. Rev. Roum. Biol.-Biol. Véget., Tome 44, No.1, 43

Hlebnikov H. A., Mogileva G.A., Rostorgueva E. Ia., 1963 – Godicinîe izmenenija okislitelino-vosstanavlivaiuşcih proţesov v pobegah hvornîh rastenii Sibiri. Trudî in-ta les i drevesini, Tom.60

Kawashima N., Uritani K., 1963 – J.Agric. Biol. Chem., Vol. 27, 409

Möller, K. M. and Ottolenghi, P., 1966 – The oxidation of o-dianisidine by H2O2 and peroxidase at neutral pH. Compts. Rend. Trav. Lab., Carlsberg, Vol.35, 369

Rubin B. A., Loginova L. H., 1965 – Fiziol. Rast., Tom 12, 386

Snedecor G. W., 1968 – Metode statistice aplicate în cercetările de agricultură şi biologie. Editura Didactică şi Pedagogică, Bucureşti, p.187

[1]„ Alexandru Ioan Cuza ” University, B-dul Carol I, 20A, 700506 Iaşi, România
* corresponding author: vartenie@uaic.ro