



Department of Floriculture and Dendrology

**CLIMATE ADAPTATION AND PEROXIDASE
ACTIVITY OF NEWLY BRED BALCONY PLANTS**

DOCTORATE THESES

CANDIDATE: SZABOLCS FEKETE

Committee members:

MRS. TILLY DR. ANDREA MÁNDY
MRS STEFANOVITS DR. ÉVA BÁNYAI

Budapest, 2008

PhD School

Name: Doctoral School of Horticultura Sciences

Field: Plant Production and Horticultural Sciences

Head of the Ph.D. School Prof. Dr. Magdolna Tóth
University Professor
Doctor of the Hungarian Academy of Sciences
Corvinus University Budapest
Faculty of Horticultural Sciences

Supervisor 1: Mrs. Tilly Dr. Andrea Mándy
Associate professor, CSc.
Corvinus University Budapest
Faculty of Horticultural Sciences

Supervisor 2: Mrs. Stefanovits Dr. Éva Bányai
Doctor of the Hungarian Academy of Sciences
Corvinus University Budapest
Faculty of Food Sciences

The applicant met the requirement of th Ph.D. regulations of the Corvinus University Budapest and the thesis is accepted for the defence process.

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Prof. Dr. Magdolna Tóth
Head of the Ph.D. School

.....
Mrs. Tilly Dr. Andrea Mándy
Mrs. Stefanovits Dr. Éva Bányai
Supervisors

1.1. INTRODUCTION

Annual plants and balcony plants among them, play a crucial role in ornamental plant production and –commerce. The numerous group of balcony plants have been discovered both by the producers and by the dealer community in the last decade. Besides the traditionally world wide painted geraniums several species were brought into production, thus practically there is a renaissance in the breeding and usage of balcony plants.

There is a need for new varieties and species besides the traditional ones and breeders attempt to fulfill these needs. Public park-maintaining companies, commercial organisations and institutions dealing with horticultural education have a major part in the evaluation of the novelty. However, in abroad, show gardens, international species-comparison institutes (AAS, Fleuroselect) also do the comparison of varieties besides the above mentioned ones.

In Hungary the popular „flower” movements for the decorating of villages and towns gave buoyancy to the application of annual ornamental plants the spreading of which is mostly done by propagation trader companies. These institutions mostly show the plants in the seedling stage, although users are also interested in the development and the vitality of those plants after plantation. Furthermore traders only exhibit the species groups to be introduced or the leader varieties.

The demonstration of the ornamental values of balcony plants is only realised through the few summer shows, but professionals still cannot meet most of the novelty, or the older thus less applied plant species.

2. AIMS

I attempted to fulfill the following major aims with my research:

- Selecting potential candidates from the yearly broadened supply of balcony plants the ones that are possibly able to adopt the extreme Hungarian climate and at the same time have satisfying ornamental value.
- Investigation of several characteristic quality traits of the chosen varieties in different locations and supplying basic data about their growth intensity and flowering characteristics.
- Identifying the usability of the plants as balcony plants based on the parameters investigated.
- Identifying the applicability of the yet not used Hungarian-bred varieties as balcony plants through planting them into window-boxes.
- Following the changes of peroxidase (POD) stress enzymes and the exploration of the connection of effects of climatic or other stress sources with this enzyme group.
- Supplying data on POD activity in the propagation material of cutting propagated ornamental plants applicable as balcony plants.

3. Materials and methods

Research locations

I chose the location according to the variable climatic traits (Figure 1). I conducted the research in the roof garden of Budai Arborétum, in Nyergesújfalu in Kisalföldön, in Kerek-hegy in Solymár and in the Great Plain, in Tázlár.



Figure 1. Research locations in Hungary
(self made map based on <http://www.budapest-geo.hu>)

The investigated plants are found in Table 1. The dark quadrangles besides the species show the experiment in which they were examined.

Table 1. Ranking of plants according to their ways of investigation in the thesis.

Latin name of the experimental plant	<i>Pre experiment carried out with Biomit Plusz</i>	<i>Rooting experiment</i>	<i>Variety evaluation and POD measurement</i>		
	2000	2002	2002	2003	2004
<i>Alternanthera ficoidea</i> 'Red'					
<i>Bidens ferulifolia</i> 'Kobold'					
<i>Bidens ferulifolia</i> 'Marigold'					
<i>Celosia argentea</i> var. <i>plumosa</i> 'Savaria'					
<i>Convolvulus sabatius</i> 'Blau Mauritius'					
<i>Dianthus chinensis</i> 'Corona Cherry Magic'					
<i>Dianthus chinensis</i> 'Super Parfait Raspberry'					
<i>Diascia barberae</i> 'Ascote Apricote'					
<i>Dichondra repens</i> 'Silver Falls'					
<i>Helichrysum bracteatum</i> 'Chico Red'					

1. táblázat. Az értekezésben vizsgált növények besorolása a kísérletek szerint - folytatás

Latin name of the experimental plant	Pre experiment carried out with Biomit Plusz	Rooting experiment	Variety evaluation and POD measurement		
	2000	2002	2002	2003	2004
<i>Iresine herbstii</i> 'Purple Lady'					
<i>Iresine lindenii</i>					
<i>Lamium</i> sp.					
<i>Lantana montevidensis</i> , White flower					
<i>Lavandula angustifolia</i>					
<i>Lobelia erinus</i> 'Fountain Lilac'					
<i>Mentha suaveolens</i> 'Bicolor'					
<i>Nemesia</i> 'Blue Bird'					
<i>Ocimum basilicum</i> 'Bíborfelhő'					
<i>Pelargonium zonale</i> 'Magic Rose'					
<i>Petunia</i> 'Million Bells Cherry'					
<i>Petunia</i> 'Million Bells Orange Scarlet'					
<i>Petunia</i> 'Easy Wave Pink'					
<i>Pilea microphylla</i>					
<i>Rudbeckia hirta</i> 'Toto Lemon'					
<i>Salvia farinacea</i> , Short, blue flower					
<i>Salvia officinalis</i>					
<i>Salvia officinalis</i> 'Purpurascens'					
<i>Santolina chamaecyparissus</i>					
<i>Sanvitalia procumbens</i> 'Aztekengold'					
<i>Sanvitalia procumbens</i> 'Orange Sprite'					
<i>Solenostemon scutellarioides</i> 'Black Dragon'					
<i>Solenostemon scutellarioides</i> 'Fantasy'					
<i>Tagetes tenuifolia</i> , Orange flower					
<i>Thymus vulgaris</i>					
<i>Tithonia rotundifolia</i> 'Narancsszőnyeg'					
<i>Verbena</i> 'Babylon Light Blue'					
<i>Verbena</i> 'Temari Scarlet'					
<i>Zinnia elegans</i> 'Zinnita Yellow'					

Laboratorial processing of the samples, POD enzyme activity measurement

For the measurements of POD enzyme activity, 1 mg of leaves and different parts of cuttings was ground in pre-cooled mortar with 1 ml ice-cold Sodium acetate buffer (Na-acetate: 0.1 M, pH=5.0). The buffer contained 10 mg/ml polivinilpirrolidone, 200 mg/ml sucrose, 0.35 mg/ml bovine serum albumine (BSA) and 100 mg/ml TritonX-100. The homogenised samples were centrifuged: 13000 rpm, 4 °C, 15 min. further analyses was carried out of the supernatant.

The POD enzyme activity was photometrically determined on a Varian DMS 100 UV-VIS spectrophotometer ($\lambda=460$ nm wave length) in the presence of H₂O₂ substrate and ortodianizidine cromogen reagent ($\epsilon=11,3$). Protein content determination (mg/ml) was carried out with the method of Bradford, and then the enzyme activity was determined for U/mg protein.

The POD enzyme activity of the samples was plotted with the help of a Microsoft Excel (XP) program, then the values read on the plot were visualised on graphs according to the sampling dates. The significance level of the samples was determined with two-templated t-test, ont he 90 % significance level.

3.1. Pre-experiment with Biomit Plussz nutrient

5 control and 5 treated boxes were placed in Nyergesújfalu and in Tázlár respectively. Control boxes were watered weekly with 2 ‰ Potassium dominant Kristalon fertiliser, the treated ones were also watered weekly with 2 ‰ of Biomit Plussz conditioner.

Sampling was made in every second week from all of the treatments, from 3 normal of the youngest leaves. Samples contained at least 5 leaves. I recorded the vitality of plants at sampling times. I covered the picked plant parts by aluminium foil, signed them and kept them in a freezer (-18 °C) until the measurements.

3.2. Rooting experiment

POD enzyme activity of balcony plants propagated from cuttings was measured in 2002. 14 ornamental plant propagated from cutting was measured.

I prepared 3-3 average samples of 5 cuttings of each taxa. Leaves and shoot ends were taken from the upper parts of the cuttings (in case of *Santolina*, *Thymus* for example). The middle part, the lower part of the shoots, and the later developed roots gave separate samples. I processed the samples in laboratory and measured their POD enzyme activity.

3.3. Evaluation of varieties

I planted the promising annual ornamental plants into window boxes in 2002, in 2003 and in 2004. In 2002 I planted 11 plant species into window boxes, and 9 in 2003 and 2004 respectively. I placed 10 window boxes in three locations. The experimental material was chosen from Hungarian, German (Kiepenkel -Nebelung) and American PanAmerican Seed.

During the evaluation I collected and evaluated those numeric morphological traits of the plants that are characteristic for the habit of the plants. I recorded the height, the width parallel and perpendicular to the window box (cm). The multiplication of the two latter data gave the horizontal spread of the plants. In the case of bagging plants I determined the extent of bagging in cm. I also recorded the flowering date and the numbers of flowers in the stage of flowering; in some cases I also recorded the sprouts and the bloomed flowers. Together with the above mentioned I furthermore recorded the vitality of the plants.

3.4. POD enzyme activity measurements during variety evaluation

I took the leaf samples in the same time of the variety evaluations from the same individuals. The data resulted from biochemical measurements were plotted on graphs.....

I plotted. During evaluation of the varieties, symptoms and the results of other observations were compared to the POD enzyme activity values.

4. Results

4.1. Pre-experiment with Biomit Plusz

In the samples taken from the leaves of *Verbena* L. 'Temari Scarlet' and of *Petunia* Juss. 'Million Bells Cherry' POD enzyme activity rose constantly. There were significant differences between the POD activity of the two varieties, resulting from different traits of the different varieties.

The enzyme activity in the initial, young period of *Verbena* L. rose to 4,8 times of the initial values. The Biomit Plusz treatment resulted in a lower, 4 times rise. The enzyme activity rise of the control was huge (0,4→1,7 U/mg protein), whereas the treated plants did not gave significant enzyme activity change (0,6→0,4 U/mg protein).

4.2. Rooting experiment

Callose tissue was formed on several taxa (e.g. *Salvia officinalis* L.) during rooting. The roots emerged in all of the plants at the final sampling time.

The POD enzyme activity was always the highest at the place of rooting, in the lower part of the cutting, the extent of which varied according to taxa. There was a decreasing tendency in the POD enzyme activity from the basal part of the cutting to the upper part.

During the rooting of *Solenostemon scutellarioides* (L.) Codd 'Fantasy' the enzyme activity was the highest before the initiation of the roots. The POD activity values rose by 4-5 times in the lower part of the cuttings. The enzyme activity of the leaves of *Aternanthera ficoidea* (L.) R. Br. ex. Roem. et Schult. 'Red' rose during rooting, but after the formation of the roots it decreased. The POD enzyme

activity of all parts of the cuttings of *Pilea microphylla* (L.) Liebm. rapidly rose up to 3-11 times of the initial values after the first few days of plantation. As the time of rooting was approached the data showed a constantly decreasing tendency in the middle and upper part of the cutting, while the POD activity began to decrease together with the emergence of the roots in the lower part of the cuttings.

Compared to the basic species of *Salvia officinalis*, *Salvia officinalis* L. 'Purpurascens' had higher POD enzyme activities (3 times) during rooting. In the case of the small leaved, branching cutting with dwarf shoots (e.g. *Thymus vulgaris* L. and *Lavandula angustifolia* Mill.) the highest enzyme activity values were not measured in the lower, but in the middle part of the cutting in the case of *Lavandula*, or in the upper or lower part of the cutting as in the case of the *Lavandula*.

4.3. Variety evaluation

The following varieties developed and ornamented constantly during vegetation period according to the results of the 27 investigated taxa, in the three years of the experiments in terms of their flowering, size and vitality results.:

- *Bidens ferulifolia* (Jacq.) DC. 'Kobold',
- *Celosia argentea* L. var. *plumosa* 'Savaria',
- *Dianthus chinensis* L. 'Corona Cherry Magic',
- *Dianthus chinensis* L. 'Super Parfait Raspberry',
- *Dichondra repens* J.R. Frost et G. Frost 'Silver Falls',
- *Iresine herbstii* Hook. f. 'Lady in Red',
- *Lantana montevidensis* (Spreng.) Briq. White flower,
- *Pelargonium zonale* (L.) L'Hér. Ex Aiton 'Magic Rose',
- *Petunia* Juss. hibrid 'Easy Wave Rose',

- *Sanvitalia procumbens* Lam. 'Aztekengold',
- *Sanvitalia procumbens* Lam. 'Orange Sprite',
- *Verbena* L. hibrid 'Babylon Light Blue'.

4.4. POD enzyme activity measurements during variety evaluation

POD enzyme activity constantly rose in case of all investigated plants throughout the whole vegetation period. After the placement of the window boxes (first date) the tendencies of POD activities were the same at all three locations, with different climatic conditions for some time (1-4 measuring date, 2-8 weeks). From that time on the pattern of POD activity changed.

Petunia Juss. 'Million Bells Orange Scarlet' showed a much less vigour and ornamental value, than 'Million Bells Cherry'. Although at the beginning of the experiment, the enzyme activities of the two varieties were similar, for the last sampling time the more sensitive species showed 1.5 time higher enzyme activity values than the variety with the stronger vigour.

Plants were subjected to several different climatic and biotic stress agents. Hot weather and the strong irradiation resulted in dropping growth or fewer flowers, both of which had an effect on POD enzyme activity. In the same time of the fewer flowers and growth rate, of *Bidens ferulifolia* (Jacq.) DC. 'Kobold', *Diascia barbarae* L. 'Ascote Apricote', *Sanvitalia procumbens* Lam. 'Aztekengold', *Lobelia erinus* L. 'Fountain Lilac' and *Verbena* L. 'Temari Scarlet' the weather was hot in the experimental locations. The decreasing tendency of growth was also followed by increased POD enzyme activity values.

Solenostemon scutellarioides (L.) Codd 'Black Dragon' showed the symptoms of sunburn, parallel with the rise in POD values.

Celosia argentea L. var. *plumosa* 'Savaria' got a severe wind effect in 2002, the consequence of which all the upright plants layed

from the window boxes and the roots of which were damaged. The POD enzyme activity of the variety rose but when the plant started to initiate new shoots, POD activity decreased to the level before the beginning of growth.

I recorded Powdery mildew in *Tithonia rotundifolia* (Mill.) S.F. Blake 'Narancsszőnyeg' növényeken, in the white flowery clone of *Lantana montevidensis* (Spreng.) Briq. and in *Petunia* L. 'Million Bells Cherry' I recorded *Helicoverpa armigera* Hbn. (Cotton bodworm) damage. POD enzyme activity rise was seen parallel with the damage in all three taxa.

For certain plants (e.g. *Tagetes tenuifolia* Cav. narancssárga virágszínű klón, *Convolvulus sabatius* Viv. 'Blaue Mauritius') biochemical measurements did not give results for proper evaluation.

NEW SCIENTIFIC RESULTS

1. I tested 27 ornamental plant species in window boxes, in three locations with different climatic conditions in the period of 2002-2004. In Hungary I evaluated *Celosia argentea* L. var. *plumosa* 'Savaria', *Dianthus chinensis* L. 'Corona Cherry Magic', *Dianthus chinensis* L. 'Super Parfait Raspberry', *Dichondra* J.R. Frost et G. Frost *argentea* 'Silver Falls', *Iresine herbstii* Hook. f. 'Purple Lady', *Lantana montevidensis* (Spreng.) Briq. White flower, *Lobelia erinus* L. 'Fountain Lilac', az *Ocimum basilicum* L. 'Bíborfelhő', *Pelargonium zonale* (L.) L'Hér. Ex Aiton 'Magic Rose', a *Petunia* Juss. hibrid 'Easy Wave Pink', *Rudbeckia hirta* L. 'Toto Lemon', *Sanvitalia procumbens* Lam. 'Orange Sprite', *Solenostemon scutellarioides* (L.) Codd 'Black Dragon', *Tithonia rotundifolia* (Mill.) S.F. Blake 'Narancsszőnyeg' and *Zinnia elegans* Jacq. 'Zinnita Yellow' taxa for the first time in terms of potential balcony application.
2. During the evaluation of the taxa I concluded that 9 had an excellent bloom: *Bidens ferulifolia* (Jacq.) DC. 'Kobold', *Celosia argentea* L. var. *plumosa* 'Savaria', *Dianthus chinensis* L. 'Corona Cherry Magic', *Dianthus chinensis* L. 'Super Parfait Raspberry', *Dichondra repens* J.R. Frost et G. Frost 'Silver Falls', *Iresine herbstii* Hook. f. 'Lady in Red', *Lantana montevidensis* (Spreng.) Briq. White flower, *Pelargonium zonale* (L.) L'Hér. Ex Aiton 'Magic Rose', *Petunia* Juss. 'Easy Wave Rose', *Sanvitalia procumbens* Lam. 'Aztekengold', *Sanvitalia procumbens* Lam. 'Orange Sprite' and *Verbena* L. 'Babylon Light Blue'.
3. I demonstrated for the first time, that the POD enzyme activity constantly rises in the leaves of the investigated balcony plants (27 taxa).

4. The seedling from the same taxa developed differently in different locations. Laboratory measurements proved that the POD activity pattern was also different.
5. I proved that the POD activity of the 'Million Bells Cherry' variety of the genus *Petunia* L. during its vegetation period rose less than the variety of 'Million Bells Orange Scarlet'. This is in accordance with the climatic sensitivity of certain varieties.
6. I concluded during the measurement of POD enzyme activity changes in case of *Verbena* L. 'Temari Scarlet' that as a consequence of the strong chorching symptoms there was a large enzyme activity rise in the control ones, while in the plants treated with the plant conditioning agent, BIOMIT PLUSSZ, the rate of enzyme activity did not change significantly. I provided data with these result of the stress resistance enhancing effect of Biomit Plussz.
7. I proved that balcony plants react with POD enzyme activity rise for abiotic stresses (high temperature, sunburn, wind damage).
8. I proved that heavy biotic stresses (Powdery mildew, Cotton bollworm) induce POD enzyme activity rise in balcony plants.
9. I proved in 14 ornamental plant taxa that after making the cuttings the POD enzyme activity rise in all parts of the cuttings, then alongside with the development of the roots, the enzyme activity drops back to the level identified at the time of the cutting making. The rise in the enzyme activity was different according to the taxa.

10. I concluded that the highest rate of enzyme activity was found during rooting, always in the location of the rooting among the investigated taxa. In some plants (e.g. *Solenostemon scutellarioides* (L.) Codd 'Fantasy' and *Pilea microphylla* (L.) Liebm.) the activity of POD rose sharply, then later, after the root development, the POD activity started to decrease in the lower part of the cutting. In the case of the small leaved, branching cutting with dwarf shoots (e.g. *Thymus vulgaris* L. and *Lavandula angustifolia* Mill.) the highest enzyme activity values were not measured in the lower, but in the middle or upper part of the cutting.
11. I observed that the POD activity of *Salvia officinalis* L. 'Purpurascens' was three times higher than the basic species, resulting from the more resistant genetic type.

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[2] Szántó M. és **Fekete Sz.** (2004): Balkonládák és függőcserepek. in Schmidt G.: Növények a kertépítészetben, Mezőgazda Kiadó, Budapest, ISBN 963 286 062 4, pp: 267-273.

Other publications beyond the topic of the thesis

Other publications:

[1] Fekete Sz. (2001): Kora tavaszi szabadföldi dísznövényeink. Őstermelő 1., pp: 60-61.

[2] Fekete Sz. (2001): Rózsató termesztése. Őstermelő 4., pp: 63-64.

[3] Fekete Sz. (2001): Cserepes krizantém termesztése. Őstermelő 5., pp: 31-32.

[4] Fekete Sz. (2001): Korán virágzó fás szárú dísznövényeink. Őstermelő 6., pp: 91-92.

[5] Fekete Sz. (2002): A vágott szegfű termesztése. Őstermelő 3., pp: 22-23.

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[7] Tillyné dr. Mándy Andrea, Fekete Szabolcs (2003): Egynyári és balkonnövény újdonságok tesztelése Magyarországon. Dísznövény Szemle, Dísznövénytermesztési és Dendrológiai Tanszék, Budapest, 2. évf. pp: 4., 6.

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[1] Fekete Sz., Tillyné Mándy A., Zsigó G. (2000): Az Osmocote Exact hatásának vizsgálata palántanevelés és virágládakiültetés estében. Lippay János – Vas Károly Tudományos Ülésszak, Dísznövénytermesztés II., Üvegházi Termesztés Szekció, pp: 108-109.

[2] Fekete Sz. (2000): Az Osmocote Exact hatásának vizsgálata balkonnövények palántanevelése és virágládákba ültetése esetében. TDK dolgozat

[3] Fekete Sz., Szurovetz D., Mándy Andrea (2003): Élő növények fejlődése és téltűrése arid viszonyok között – Entwicklung und Wintertoleranz von mehrjährigen Zierpflanzen unter Trockenbedingungen. Lippay János – Ormos Imre – Vas Károly Tudományos Ülésszak, Összefoglalók, Kertészettudomány, pp: 192-193.