

EFFECT OF METHYL JASMONATE VAPORS ON LEVEL OF ANTHOCYANINS, BIOGENIC AMINES AND DECARBOXYLASES ACTIVITY IN SEEDLINGS OF CHOSEN VEGETABLE SPECIES

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Abstract. Seedlings of four vegetable species (maize, tomato, radish and onion) were treated for 7 days with methyl jasmonate (MeJA) vapors. MeJA accelerated senescence process of plant tissues and accumulation of anthocyanins. The only exception were hypocotyls of radish, which was found to decrease of the anthocyanin content under the influence of MeJA. It has been shown that MeJA has a different impact on the content of free biogenic amines. In case of leaves and epicotyls of maize and tomato hypocotyls, MeJA had no effect on levels of putrescine (Put). The leaves of tomatoes have shown to increase the putrescine level as a result of the impact of MeJA vapors. However, in the tissues of radish and onion very large decline in putrescine and spermidine content under the influence of the phytohormone were observed. The presence of small amounts of spermine was found only in tissues of radish, and onion, which does not affect the use of MeJA. In tissues of maize the presence of a significant content of 2-phenylethylamine (PEA) were found. Use for 7 days of MeJA vapors resulted in 3-fold increase in the content of the PEA in maize leaves. Small levels of the amine has also been found in tomato hypocotyls, where use of MeJA caused reduction its content. Obtained results show a little relationship between the activity of the studied enzymes (ornithine decarboxylase (ODC), lysine decarboxylase (LDC) and tyrosine decarboxylase (TYDC)) and the contents of the amines in seedlings of four vegetable species. No free cadaverine and tyramine were found, and therefore probably both polyamines might be present as conjugates. Putrescine also may be present in bound form. Moreover, since putrescine can be synthesized directly from ornithine or indirectly from arginine *via* agmatine, the activity of ODC alone did not give a full picture of the impact of MeJA on Put accumulation.

Key words: methyl jasmonate, seedlings, anthocyanins, amines, decarboxylases

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INTRODUCTION

Jasmonic acid (JA), its methyl ester (MeJA) and other conjugates called jasmonates, are compounds widely distributed in the plant kingdom [Wasternack 2007]. JA is produced from α -linolenic acid present in chloroplast membranes. Jasmonates are involved in crucial processes related to plant development and survival, including defense responses, secondary metabolism, reproductive process, senescence and fruit development [Creelman and Mullet 1995, Farmer et al. 2003, Wasternack 2007]. Jasmonates have been shown to be powerful promoters of leaf senescence [Hung and Kao 2004, Ananieva et al. 2007]. The compounds accelerate the leaf abscission and reduce the chlorophyll level [Ueda and Kato 1981, Weidhase et al. 1987, He et al. 2002].

Methyl jasmonate has been considered, due to its volatility, as an important candidate for an airborne signal molecule mediating interplant communications and modulating plant defense responses [Creelman and Mullet 1995, Wasternack 2007, Tamogami et al. 2008].

Jasmonates affect the content and metabolism of polyamines in plant tissues. This effect varies and is dependent on species studied, the type of tissue, culture methods used and other factors. According to Mader [1999] in roots and shoots of micropropagated potato plants JA reduced free polyamines, whereas polyamine conjugates increased up to 10-fold. Application of MeJA promotes accumulation of caffeoylputrescine in tomato leaves, a metabolite, which combines a pathway of polyamines and phenylpropanoids [Chen et al. 2006]. MeJA also induced accumulation of conjugated polyamines and activity of ornithine and S-adenosylmethionine decarboxylases, but not arginine decarboxylase in tobacco leaf discs [Biondi et al. 2003]. During Biondi et al. [2001] studies MeJA declined levels of free putrescine, spermidine and spermine in tobacco thin layers after 7 and 15 days of treatment. Activity of arginine and ornithine decarboxylases displayed up to 3-fold increases relative to control explants. In case of seedlings MeJA induced the accumulation of putrescine in detached rice leaves incubated in darkness, and grown in the light conditions [Chen et al. 1994]. In another studies on rice plants MeJA induced putrescine and spermine accumulation, while spermidine levels decreased [Lee et al. 1996].

Accumulation of anthocyanins in plant seedlings is stimulated by various environmental factors, such as high-intensity light, wounding, pathogen attack, drought and nutrient deficiency [Mancinelli 1990, Rabino and Mancinelli 1986, Steyn et al. 2002]. Methyl jasmonate as a known factor in acceleration of aging of plant tissues also causes accumulation of anthocyanins [Saniewski et al. 2003, 2006, Shan et al. 2009]. However, certain plant tissues respond differently to the MeJA. Recently it was found that MeJA strongly inhibited roots elongation in buckwheat seedlings and decreased anthocyanin accumulation in hypocotyls [Horbowicz et al. 2008]. On the other hand in buckwheat tissues MeJA stimulated the biosynthesis of putrescine, 2-phenylethylamine and proanthocyanidins, but had no influence on the content of major flavonoids: glycosides of apigenine, luteoline and quercetine [Horbowicz et al. 2011a, b]. It was suggested that for the decline of anthocyanins content in buckwheat hypocotyls by MeJA may be partly responsible L-phenylalanine deficiency – a substrate for synthesis of 2-phenylethylamine (PEA), formed by decarboxylation of the amino acid [Horbowicz et al. 2013].

Aromatic amino acids are important precursors of various secondary metabolites in higher plants. Enzymes involved in amino acids transformation, such as phenylalanine ammonia lyase [Hahlbrock and Grisebach 1979], and decarboxylases are inducible by various factors [Chapple et al. 1986]. These regulatory enzymes are of particular interest due to their role in secondary metabolism in plants. Phenylalanine ammonia lyase (PAL) catalyzes the deamination of L-phenylalanine to *trans*-cinnamic acid, the first step in the biosynthesis of phenylpropanoids, coumarins, flavonoids, and lignin [Hahlbrock and Scheel 1989].

On the other hand, decarboxylation of amino acid provides a number of secondary metabolites, which are not essential for normal growth and development, but are often involved in interactions between plants and biotic and abiotic factors [Facchini et al. 2000]. Major group of such metabolites are polyamines. Polyamines are involved in a wide range of plant processes [Groppa and Benavides 2008]. Due to their role in production of hydrogen peroxide by amine oxidases, polyamines are involved in mediate of the stress responses.

The aim of this study was to compare the effect of MeJA vapors (promoter of senescence) on the accumulation of anthocyanins and polyamines, as well as the major decarboxylases activity in two species of monocotyledonous and two dicotyledonous. Due to the lack of data in the scientific literature additional aim was to determine whether formation 2-phenylethylamine (PEA) is a widespread phenomenon in plants. With the increasing consumption of sprouts due to the high content of antioxidants and vitamins seems to be important knowledge about the ingredients found in small amounts, as PEA. PEA can affect human health and well-being, and can increase blood pressure and cause migraine, therefore its level in food can be important [Shalaby 1996].

MATERIALS AND METHODS

Seedlings of radish (*Raphanus sativus* var. *sativus*, cv. Krakowianka) and maize (*Zea mays* L. subsp. *mays* Grupa Saccharata, cv. Karłowa Złota) were prepared by germination of seeds placed between two layers of wet filter paper which were then rolled and inserted in a 2.5 l beaker containing ca. 200 cm³ of tap water. The germination process was carried out in darkness at 24 ±1°C. After four days of germination the obtained seedlings were taken to experiments with methyl jasmonate vapors.

In case of tomatoes (*Lycopersicon esculentum* Mill., cv. Jontek) 4-weeks old seedlings obtained by germination and cultivation in peat soil were used, under the light and temperature conditions described below (air conditioned room). Tomato seedlings were carefully removed from the soil and placed between two layers of wet filter paper which were then rolled and inserted in beaker used for experiments. During experiments with methyl jasmonate plants of radish, maize and tomato were grown in one fifth Hoagland nutrient solution.

The leaves of onion (*Allium cepa* L.) were obtained by forcing of spring onions (cv. Wolska) bulbs for 10 days, under the light and temperature conditions described below (air conditioned room). Each bulb was placed in small (ca. 50 cm³) pot filled with peat

soil. When the leaf reached a length of 8–10 cm plants with pots were placed into beakers used to experiments with methyl jasmonate vapors.

The all seedlings and plants were exposed to methyl jasmonate vapors in 2.5 L beakers tightly closed with silicon foam, by incubating with strip of filter paper to which had been applied 50 μL of MeJA. MeJA gradually evaporated, and calculated its concentration does not exceed a level of 1 ppm. Control plants were grown in beaker closed with silicon foam, but strip of filter paper was without MeJA. One replicate contained from 20 to 40 seedlings in the beaker. To experiment for each plant 3 beakers was taken, and plants from one beaker was treated as replicate.

All the seedlings treated with MeJA and control were grown in an air conditioned room, in which the temperature was maintained at $22 \pm 2^\circ\text{C}/18 \pm 2^\circ\text{C}$ (day/night: 16 h/8 h). Light ($100 \mu\text{Mol} \times \text{m}^{-2} \times \text{s}^{-1}$) was provided by high-pressure sodium lamps. After seven days in such conditions, plants were subjected to analysis of anthocyanins, biogenic amines and decarboxylases activity.

HPLC determination of amines. Free amines were analysed according to procedure described by Flores and Galston [1982], later slightly modified [Horbowicz et al. 2008]. Briefly, fresh plant tissues (0.5 g in 4 mL) were homogenized in 5% (v/v) perchloric acid and homogenates were centrifuged, and amines were derivatised with benzoyl chloride. Benzoyl derivatives of amines were extracted with ethyl acetate, and the solvent was evaporated in stream of warm air. The residue was dissolved in a mobile phase used for HPLC analyses (acetonitrile-water, 45:55). Benzoylated amines were eluted isocratically at the temperature using a column Eclipse XDB-C18 analytical ($4.6 \times 150 \text{ mm}$, $5 \mu\text{m}$ particle size). The benzoyl derivatives were detected at 245 nm, diode array detector, and amine contents were calculated from standard curves of commercially available standards.

Determination of anthocyanins. Total anthocyanin content was analyzed spectrophotometrically using a method described by Mancinelli [1984] with some modifications [Horbowicz et al. 2008]. For one analysis was taken 0.2–0.4 g of finely chopped fresh tissue. For the measurement of the anthocyanin contents a UV-VIS spectrophotometer Hewlett Packard 8453 was applied.

Assay of the enzymes activity. Ornithine decarboxylase (ODC) activity was assayed according to Ngo et al. [1987]. Briefly, plant tissues (1.00 g) were homogenized in phosphate buffer (pH 8.2) contained β -mercaptoethanol and EDTA (10 mL). The enzyme extract was obtained by filtration through cheese cloth and centrifuged at 18 000 g at 5°C . Then the extract was mixed with ornithine in phosphate buffer (pH 8.2) contained pyridoxal-5-phosphate. After incubation at 30°C for 30 minutes the reaction was stopped by adding 10% trichloroacetic acid, 4 M NaOH and 1-pentanol. Following mixing and centrifuging separated organic layer was transferred to glass tube contained 10 mM trinitrobenzosulfonic acid in 1-pentanol, and DMSO. After vigorous mixing and centrifugation absorbance of the organic layer (putrescine) was measured at a wavelength of 426 nm.

Determination of lysine decarboxylase (LDC) activity was conducted according to Phan et al. [1982] method. Plant tissues (1 g) were homogenized in 10 mL of 0.2 M Tris-HCl buffer (pH 5.6). The enzyme extract was obtained by filtration through cheese cloth and centrifuged at 18 000 g at 5°C . Enzyme extract was mixed with 20 μM lysine

and 0.1 μM of pirydoxal-5-phosphate. The mixture was incubated at 30°C for 20 min and enzymatic reaction was stopped by adding 1 M potassium carbonate and trinitrobenzosulfonic acid. Obtained product of reaction (cadaverine) was extracted with toluene. Absorbance of the toluene layer was measured at 340 nm.

The tyrosine decarboxylase (TYDC) activity was analyzed according to method described by Phan et al. [1983]. Plant tissues (1 g) were homogenized in 10 mL of acetate buffer (pH 5.6). The enzyme extract was obtained by filtration through cheese cloth and centrifuged at 18 000 g at 5°C. Enzyme extract was mixed with tyrosine and 0.1 μM of pirydoxal-5-phosphate. Then the mixture was incubated at 30°C for 30 min and reaction was stopped by addition 1 M potassium carbonate and trinitrobenzosulfonic acid. Obtained product of reaction (tyramine) was extracted with toluene. Absorbance of the toluene layer was measured at 340 nm.

For the measurement of the activity of all enzymes a UV-VIS spectrophotometer Hewlett Packard 8453 was applied. Activity of enzymes was expressed in μM of appropriate polyamine generated during 1 hr of the enzymatic reaction by 1 mg of protein. Estimation of protein quantity within the enzymatic extracts was performed according to Lowry et al. [1951] method.

Statistical analyses. All measurements were done in three replicates. Student's *t*-test was used for statistical evaluation of the differences between the control and treated plants.

RESULTS AND DISCUSSION

Anthocyanins in plants treated with MeJA. Effect of MeJA vapors on anthocyanin accumulation was dependent on the type of tissue and tested plant (tab. 1). In most cases, MeJA caused increase in accumulation of anthocyanins. The results are mostly compatible with previous reports, which indicated that the methyl jasmonate induces the accumulation of anthocyanin in tissues of tulip stems [Saniewski et al. 1998a], peach shoots [Saniewski et al. 1998b], *Kalanchoe blossfeldiana* [Saniewski et al. 2003], and shoots of *Crassula multicaeva* [Saniewski et al. 2006]. It can be assumed that the increased accumulation of anthocyanins may be caused by MeJA as a factor stimulating senescence [Saniewski et al. 2003, 2006, Shan et al. 2009, Ueda and Kato 1981].

During the senescence process decline of chlorophyll is used to be observed. Consequently, it may increase photoinhibition phenomenon. By absorbing of excess of radiation before it reaches the chloroplasts, anthocyanins therefore have the potential to reduce the photoinhibition. This anthocyanin function has been confirmed several times in various studies [Close and Beadle 2003, Chalker-Scott 1999, Gould 2004, Steyn et al. 2002].

However, radish hypocotyls responded differently, in which methyl jasmonate caused a marked reduction (three-fold) in the level of anthocyanins, compared to untreated plants. The phenomenon of reducing the level of anthocyanins in plant tissues under the influence of MeJA is relatively rare. The only known and similar effect of MeJA on anthocyanins was found earlier in hypocotyls of common buckwheat seedlings [Horbowicz et al. 2008]. It was suggested that possibly reason of anthocyanins

decline is competition between transformation of L-phenylalanine to *trans*-cinnamic acid, and decarboxylation of the amino acid to large quantities of 2-phenylethylamine (PEA).

Table 1. Effect of 7-days treatment with vapors of methyl jasmonate on anthocyanins content ($\mu\text{g}\cdot\text{g}^{-1}$ fresh weight) in seedlings of selected vegetable species

Vegetable species		Leaves	Epicotyls
Maize	control	23.9 \pm 6.0	48.3 \pm 3.1
	MeJA treated	48.4 \pm 2.0 *	62.4 \pm 8.3
Tomato	control	304.1 \pm 18.9	93.1 \pm 3.6
	MeJA treated	644.1 \pm 9.0**	157.2 \pm 7.1**
Radish	control	41.3 \pm 2.3	225.5 \pm 23.8
	MeJA treated	312.8 \pm 26.0**	72.9 \pm 3.6**

Asterisks * and ** indicate significance of difference from control according to Student t-test with $p \leq 0.05$ and $p \leq 0.01$, respectively

Effect of MeJA on biogenic amines. Under stress, different plant species vary in their response in terms of polyamine changes. Some species might accumulate polyamines in response to stress, while others do not or even decrease polyamine level [Walters 2003]. Among the analyzed amines putrescine has occurred in the all tissues examined, and presence of spermidine was found in all plants, except for maize. The 2-phenylethylamine appeared in measurable amounts in the tissues of maize (leaves and epicotyls) and in tomato hypocotyls, and spermine in the tissues of tomato and onion leaves (tab. 2).

Lasting for 7-days treatment of plants by vapors of the MeJA has been affected in various ways on content of the amines. There was no significant effect on the level of putrescine in maize tissues and tomato hypocotyls, but in the leaves of tomato MeJA caused increase the amine content. The opposite phenomenon occurred in the tissues of radish and onion leaves, in which the methyl jasmonate decreased the putrescine content. In particular, a huge decline (approximately 10-fold) in putrescine level under the influence of MeJA took place in the leaves and hypocotyls of radish.

There are few scientific reports on the impact of MeJA on polyamine biosynthesis and metabolism in plants. Exogenous treatment with jasmonates has been shown to reduce the levels of free polyamines in some plant tissues, like potato shoot and roots [Mader 1999] and tobacco thin layers [Biondi et al. 2001], however induced accumulation of putrescine in rice leaves [Chen et al. 1994, Lee et al. 1996]. These results indicate that monocots probably react differently to jasmonates than dicotyledonous plants. Our findings seem to confirm this observation.

Recently published results have shown that methyl jasmonate increases the levels of putrescine but do not affect the content of spermidine in cotyledons of common buckwheat seedlings [Horbowicz et al. 2011a]. The present studies on the impact of MeJA on the polyamines in tomato tissue are similar to those for buckwheat. In both cases MeJA enhanced putrescine content in leaves but not in hypocotyls, and did not affect

the levels of spermidine. It has been shown that exogenous application of MeJA greatly promotes accumulation of caffeoylputrescine in tomato leaves [Chen et al. 2006]. Our results partially confirm the report. It is likely that part of putrescine in the leaves of tomato can be converted into a hydroxycinnamic acid amide, however further studies are needed in this area.

Table 2. Effect of 7-days treatment with vapors of methyl jasmonate on level of free biogenic amines (nmol·g⁻¹ fresh weight) in seedlings of selected vegetable species

		Putrescine	Spermidine	Spermine	2-Phenylethylamine (PEA)
Maize, leaves	control	381.8 ±37.4	–	–	94.1 ±21.7
	MeJA treated	463.5 ±101.3	–	–	278.2 ±45.2*
Maize, epicotyls	control	423.0 ±58.0	–	–	9.0 ±9.8
	MeJA treated	398.9 ±49.8	–	–	9.9 ±10.4
Tomato, leaves	control	88.1 ±9.9	127.7 ±7.5	17.7 ±4.4	–
	MeJA treated	176.0 ±4.0**	133.0 ±7.0	19.6 ±1.3	–
Tomato, hypocotyls	control	32.4 ±4.4	30.2 ±1.1	3.9 ±1.3	18.6 ±1.0
	MeJA treated	29.1 ±3.9	24.3 ±1.3	6.8 ±0.8	4.3 ±0.3**
Radish, leaves	control	3608.0 ±251.0	342.3 ±16.5	–	–
	MeJA treated	327.0 ±51.0**	53.1 ±13.7**	–	–
Radish, hypocotyls	control	2381.0 ±173.0	265.7 ±8.5	–	–
	MeJA treated	313.7 ±26.5**	67.0 ±8.5**	–	–
Onion, leaves	control	72.6 ±6.7	115.2 ±5.6	7.0 ±1.9	–
	MeJA treated	48.0 ±7.4*	45.8 ±7.0**	4.5 ±0.4	–

Asterisks * and ** indicate significance of difference from control according to Student t-test with $p \leq 0.05$ and $p \leq 0.01$, respectively

The huge decline in putrescine and spermidine content in leaves and hypocotyls of radish was probably caused by accelerated aging of the tissues under the influence of MeJA. There was observed a partial loss of the green color (chlorosis) in the leaves at the end of the experiment. It can also provide a great susceptibility of radish leaves to senescence. There is a lack in available scientific literature data on the impact of MeJA on level of polyamines in radish seedlings.

Relatively high amounts of PEA in the leaves of maize seedlings are also not supported by the available literature, and applied of MeJA vapors enhanced its level (tab. 2). PEA is quite rare amine in higher plants [Smith 1977, Shabana et al. 2006]. In tomato plants PEA can be transformed into 2-phenylacetaldehyde, and further converted to 2-phenylethanol [Tieman et al. 2006]. Recently was found that exogenous MeJA has a great stimulatory impact on the synthesis of PEA in hypocotyls and cotyledons of common buckwheat (*Fagopyrum esculentum* Moench). High concentration of PEA in

buckwheat tissues is probably the plant response to stimulation the senescing process by exogenous MeJA [Horbowicz et al. 2011a]. In humans excess of PEA may increase of blood pressure and cause migraine [Shalaby 1996].

Effect of MeJA on activity of some decarboxylases. Our results show little relationship between the activity of the studied enzymes and the contents of the main amines in seedlings of examined four species of vegetables. It was found the induction of ornithine decarboxylase (ODC) activity under the influence of MeJA within leaves and hypocotyls of radish and onion leaves, however, the activity was reduced within maize leaves (tab. 3). In case of lysine decarboxylase (LDC) activity increased within leaves of tomato and onion as well as radish hypocotyl and decreased in radish leaves, however, there was no presence of LDC product – cadaverine in any examined tissue (tab. 2, 3). These results indicate also that changes in the accumulation of polyamines in studied plant tissues under influence of MeJA were not associated with post translational regulation of ODC and LDC activity. We suppose that there can be three possible explanations of this phenomenon.

Firstly, changes in protein concentration in ODC extracts (data not shown) are compatible with variations in putrescine level, what may suggest that MeJA rather directly affect regulation of genes encoding enzyme proteins. According to Cheong and Choi [2003] exogenous MeJA upregulated genes involved in jasmonate biosynthesis, secon-

Table 3. Effect of 7-days treatment with vapors of methyl jasmonate on activity of some decarboxylases (expressed as μM of appropriate amine $\cdot \text{h}^{-1} \cdot \text{mg}^{-1}$ protein) in seedlings of selected vegetable species

		Ornithine decarboxylase (ODC)	Lysine decarboxylase (LDC)	Tyramine decarboxylase (TyDC)
Maize, leaves	control	423.6 \pm 13.0	30.2 \pm 1.8	0
	MeJA treated	256.7 \pm 11.5*	28.6 \pm 3.1	0
Maize, epicotyls	control	824.7 \pm 54.3	139.0 \pm 16.9	0
	MeJA treated	966.1 \pm 81.5	188.0 \pm 8.8	0
Tomato, leaves	control	142.9 \pm 5.5	25.7 \pm 3.0	173.5 \pm 7.4
	MeJA treated	140.4 \pm 16.0	33.0 \pm 2.4	158.3 \pm 15.0
Tomato, hypocotyls	control	179.0 \pm 12.4	37.0 \pm 10.1	526.4 \pm 2.4
	MeJA treated	174.1 \pm 15.3	53.7 \pm 4.1	422.5 \pm 9.8*
Radish, leaves	control	156.3 \pm 5.1	68.5 \pm 5.6	65.1 \pm 8.1
	MeJA treated	502.7 \pm 22.2**	28.0 \pm 12.0	94.1 \pm 6.4
Radish, hypocotyls	control	222.9 \pm 4.8	25.3 \pm 12.6	115.3 \pm 7.9
	MeJA treated	309.3 \pm 9.8*	107.4 \pm 13.5*	154.3 \pm 6.9*
Onion, leaves	control	859.7 \pm 7.8	15.7 \pm 1.6	54.5 \pm 20.2
	MeJA treated	1333.0 \pm 16.8**	83.4 \pm 5.5**	284.6 \pm 14.0**

Asterisks * and ** indicate significance of difference from control according to Student t-test with $p \leq 0.05$ and $p \leq 0.01$, respectively

dary metabolism, cell-wall formation and those encoding stress-protective and defensive proteins. In case of *Theobroma cacao* MeJA induced gene of class VII chitinase and caffeine synthase in young red leaves, and type III peroxidase in mature green [Bailey et al. 2005]. According to Xu et al. [2004] ODC transcript levels increased within transgenic tobacco cells following treatment with MeJA. However, in case of transgenic rice exogenous MeJA transiently inhibited expression of genes encoding such enzymes of polyamine biosynthesis as: arginine decarboxylase, S-adenosylmethionine decarboxylase and spermidine synthase [Peremarti et al. 2010]. Our results suggest that the MeJA upregulated genes encoding ODC and LDC in leaves of maize and tomato, but down-regulated in other plant tissues, which was particularly evident in the case of the leaves of radish. However, this hypothesis should be verified by a more detailed study.

Secondly, within plant cells putrescine can be synthesized directly from ornithine, or indirectly from arginine *via* agmatine by arginine decarboxylase (ADC) and participation of two appropriate hydrolases: agmatine iminohydrolase and *N*-carbamoylputrescine amidohydrolase [Walters 2003]. It seems like in case of some plants (radish and onion) putrescine is partly produced by indirect pathway. The MeJA likely strongly inhibit the activity of one or more enzymes responsible for this type of the putrescine synthesis. Moreover, level of free polyamines may be also regulated through their degradation. Thus, the next studies should be focused on studies of MeJA influence on ADC activity as well as enzymes catalyzed oxidative degradation of these compounds such as diamine oxidase and polyamine oxidase within examined plants.

Thirdly, considerable part of plant amines is not accumulated in free form, but as hydroxycinnamic acid amides (HCAAs). HCAAs are main low molecular weight phenolic components of generative organs in about 20 plant species of various families [Walters 2003]. The HCAAs level can be controlled by changes in activity of key enzymes in polyamine biosynthesis. This conclusion confirms the results of previous studies in which induction of ODC and ADC in MeJA-treated plants was accompanied by a decrease of free polyamines, and a significant increase of the HCAAs level has been confirmed [Mader 1999, Biondi et al. 2001, 2003]. This can be also explained by presence of an active LDC or TYDC and their changes, which may potentially suggests a connection with pool of HCAAs or other derivatives, but not free polyamines, since free cadaverine and tyramine were not detected in analyzed plant tissues.

CONCLUSIONS

Methyl jasmonate (MeJA) vapors used for 7 days accelerated accumulation of anthocyanins in tissues of vegetable seedlings: maize, tomato, radish and onion. The only exception were hypocotyls of radish, which was found to decrease of the anthocyanin content. It has been shown that MeJA has a different impact on the content of free biogenic amines. MeJA had no effect on levels of putrescine in leaves and epicotyls of maize and tomato hypocotyls, but leaves of tomatoes have shown to increase the amine level. In case of radish and onion tissues a very large decline in putrescine and spermidine content under the influence of the MeJA were observed. Use MeJA vapors resulted in 3-fold increase in the content of the 2-phenylethylamine in maize leaves. Ob-

tained results show a little relationship between the activity of the studied enzymes (ornithine decarboxylase, lysine decarboxylase and tyrosine decarboxylase) and the contents of the amines in seedlings of four vegetable species.

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WPLYW PAR JASMONIANU METYLU NA POZIOM ANTOCYJANÓW, AMIN BIOGENNYCH I AKTYWNOŚCI DEKARBOKSYLAZ W SIEWKACH WYBRANYCH GATUNKÓW WARZYW

Streszczenie. Siewki czterech gatunków warzyw (kukurydzy, pomidora, rzodkiewki i cebuli) traktowano przez 7 dni parami jasmonianu metylu (MeJA). MeJA przyspieszył proces starzenia tkanek roślinnych, powodując nagromadzenie antocyjanów. Jedynym wyjątkiem był hipokotyle siewek rzodkiewki, w których stwierdzono zmniejszenie zawartości antocyjanów pod wpływem par MeJA. Wykazano, że MeJA ma zróżnicowany wpływ na zawartość amin biogennych. W przypadku liści i epikotyli kukurydzy oraz hipokotyli pomidora pary MeJA nie miały wpływu na zawartość putrescyny (Put). Z kolei

liście pomidorów reagowały wzrostem poziomu putrescyny pod wpływem par MeJA. Natomiast w tkankach cebuli i rzodkiewki obserwowano bardzo duży spadek zawartości Put i spermidyny pod wpływem tego fitohormonu. Stwierdzone w tkankach rzodkiewki i cebuli niewielkie zawartości sperminy nie ulegały zmianom pod wpływem par MeJA. W tkankach kukurydzy wykazano obecność znacznej zawartości 2-fenyletyloaminy (PEA). Stosowanie przez 7 dni par MeJA spowodowało 3-krotny wzrost zawartości PEA w liściach siewek tego gatunku. Niewielki poziom PEA stwierdzony w hipokotylach pomidorów ulegał obniżeniu pod wpływem MeJA. Uzyskane wyniki wskazują na mały związek pomiędzy aktywnością badanych enzymów (dekarboksylazy ornityny – ODC, dekarboksylazy lizyny – LDC i dekarboksylazy tyrozyny – TyDC) i zawartości amin w siewkach czterech badanych gatunków warzyw. W tkankach analizowanych gatunków warzyw nie stwierdzono mierzalnych poziomów kadaweryny i tyraminy, co może sugerować, że obie aminy mogą występować wyłącznie w formie związanej. Ponieważ putrescyna może być syntetyzowana bezpośrednio z ornityny lub pośrednio z argininy poprzez agmatynę, aktywność ODC nie daje pełnego obrazu wpływu MeJA na jej nagromadzenie.

Słowa kluczowe: jasmonian metylu, siewki, antocyjany, aminy, dekarboksylazy

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