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Fatty acid and sterol contents during methyl jasmonate-induced leaf abscission in *Kalanchoe blossfeldiana*

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Abstract

It was found previously that methyl jasmonate (JA-Me) induced leaf abscission in *Kalanchoe blossfeldiana*. In present studies it was showed that JA-Me did not affect or only slightly affected the content of free and bound fatty acids in petioles and blades. ß-Sitosterol, campesterol and ß-amyrin were identified in petioles and blades of *K. blossfeldiana*; JA-Me decreased the content of campesterol in petioles and increased the content of ß-sitosterol in blades. In blades of plants treated with JA-Me disappearance of olean-12-one was indicated but appearance of 2H-cyclopropa[a]-naphthalen-2-one,1, 1a, 4, 5, 6, 7, 7a, 7b-octahydro-1, 1, 7, 7a-tetramethyl (aristolone) was documented. The significance of these findings in leaf abscission induced by methyl jasmonate in *K. blossfeldiana* is discussed.

INTRODUCTION

Many studies concerned the role of jasmonic acid (JA) and methyl jasmonate (JA-Me) in senescence of different organs, mostly leaves (U e d a and K a t o, 1980), but only a little attention has been paid to the role of the hormones in abscission of leaves and other organs. U e d a et al. (1991) found that quantitative changes of abscisic acid and methyl jasmonate correlated with vernal leaf abscission of *Ficus superba* var. *japonica*. S a n i e w s k i and W ę g r z y n o w i c z - L e s i a k (1994) showed that JA-Me greatly promoted the leaf abscission in intact *Kalanchoe blossfeldiana* plants treated before flower bud initiation and during flowering. The leaf abscission induced by methyl jasmonate in *Kalanchoe blossfeldiana* is not related to ethylene production.

Methyl jasmonate action is related with lipid metabolism. It was found recently that methyl jasmonate induced the expression and accumulation of lipoxygenase activity in seedlings and suspension culture of soybean (K a t o et al., 1993;

Grimes et al., 1992; Bell and Mullet, 1991; Park et al., 1994). Ranjan and Lewak (1992) reported that methyl jasmonate greatly stimulated lipase activity in non-stratified apple embryos and this way mobilized the lipid reserves.

In this study the effect of methyl jasmonate on fatty and sterol contents during induced leaf abscission in *Kalanchoe blossfeldiana* were analyzed.

MATERIAL AND METHODS

Kalanchoe blossfeldiana Poelln., a crassulacean acid metabolism (CAM) plant (Brulfert et al., 1993), is a short day plant. Cuttings (young shoots) were taken from intact *Kalanchoe* at the end of April and were rooted in the medium consisted of soil, peat moss and sand, under natural glasshouse conditions. After 1 month, the well rooted cuttings were planted individually in pots and cultivated according to standard conditions.

At the end of October and on the beginning of February the plants were sprayed with methyl jasmonate solution at a concentration of 800 mg/l in 0.05 % Tween 80. Plants treated with 0.05 % Tween 80 were used as a control. Six to ten plants were used per treatment and 30 ml of solution per plant were used for spraying. One day after treatment the petioles and leaf blades from 50 leaves were collected and frozen at -20°C. Frozen samples were used for analysis of fatty acid and sterol contents.

Analysis of fatty acid and sterol contents were previously described (Saniewski et al., 1994). Three unknown peaks were identified as: ß-amyrin (match quality 90 %), olean-12-one (match quality 80 %) and 2H-cyclopropa[a]napthalen-2-one, 1, 1a, 4, 5, 6, 7, 7a, 7b-octahydro-1, 1, 7, 7a-tetramethyl (match quality 93 %) using references mass spectrum library NBS49K.

Results were subjected to an analysis of variance and evaluated using t-test at 5 % level of significance.

RESULTS AND DISCUSSION

Spraying of intact *Kalanchoe blossfeldiana* with methyl jasmonate at a concentration of 800 mg/l induced all leaves abscission two days after treatment. The all analyses of fatty acids and sterols content were made one day after treatment with JA-Me when the leaves easy abscisse after gentle shaking.

The content of free fatty acids in petioles and blades of *Kalanchoe blossfeldiana* is very low in both the samples taken on October and February (Tab. 1 and 2). Methyl jasmonate significantly lowered the content of fatty acids in blade of plants treated in October (Tab. 1).

Both in petioles and blades in highest concentrations occurred linolenic, linoleic and palmitic acids, independently from the stage of development (Tab. 1 and 2). Methyl jasmonate did not affect or only slightly affected the content of bound fatty acids (Tab. 1 and 2).

Table 1

The effect of methyl jasmonate on the content of some free and bound fatty acids (μg g^{-1} fresh weight) in petioles and blades of *Kalanchoe blossfeldiana* 1 day after treatment (October 25) before flowering

Fatty acids	Petioles			Blades		
	Control	JA-Me	$LSD_{0.05}$	Control	JA-Me	$LSD_{0.05}$
	Free fatty acids					
Myristic (C 14:0)	< 1.0	< 1.0	NS	16.1a	7.1b	3.6
Palmitic (C 16:0)	3.5a	2.5b	0.6	35.3a	17.4b	9.0
Palmitoleic (C 16:1)	1.6	1.4	Ns	–	–	–
Stearic (C 18:0)	2.5	2.3	NS	4.9a	3.7b	1.0
Oleic (C 18:1)	2.1	2.2	NS	7.6a	4.9b	2.3
Linoleic (C 18:2)	2.1	2.2	NS	28.0a	18.2b	5.7
Linolenic (C 18:3)	< 1.0	< 1.0	NS	52.7a	34.5b	12.3
	Bound fatty acids					
Myristic (C 14:0)	18b	28a	8	13b	30a	14
Palmitic (C 16:0)	239	218	NS	281	251	NS
Palmitoleic (C 16:1)	23b	49a	14	41	49	NS
Stearic (C 18:0)	69	60	NS	75	89	NS
Oleic (C 18:1)	72b	117a	43	103	112	NS
Linoleic (C 18:2)	670	842	NS	502	524	NS
Linolenic (C 18:3)	661b	1043a	223	1221b	1854a	553

Table 2

The effect of methyl jasmonate on the content of some free and bound fatty acids (μg g^{-1} fresh weight) in petioles and blades of *Kalanchoe blossfeldiana* 1 day after treatment (February 7) during full flowering

Fatty acids	Petioles		Blades		$LSD_{0.05}$
	Control	JA-Me	Control	JA-Me	
	Free fatty acids				
Myristic (C 14:0)	2.4b	1.5b	4.0a	4.8a	1.3
Palmitic (C 16:0)	7.9b	6.2b	14.1a	16.4a	4.2
Palmitoleic (C 16:1)	3.4b	1.6b	6.2a	9.8a	2.7
Stearic (C 18:0)	1.4	1.1	1.7	1.8	NS
Oleic (C 18:1)	4.1b	3.3b	7.3a	8.6a	3.0
Linoleic (C 18:2)	2.8b	3.6b	7.2a	6.3a	1.8
Linolenic (C 18:3)	3.2c	4.9c	8.3b	13.6a	2.5
	Bound fatty acids				
Myristic (C 14:0)	41b	38b	48a	50a	4.0
Palmitic (C 16:0)	255bc	251c	273ab	292a	22.0
Palmitoleic (C 16:1)	19	20	20	18	NS
Stearic (C 18:0)	53	56	54	50	NS
Oleic (C 18:1)	47c	64a	51bc	58ab	8.0
Linoleic (C 18:2)	439a	441a	370b	425a	35.0
Linolenic (C 18:3)	281c	297c	668b	792a	65.0

ß-Sitosterol, campesterol and ß-amyrin (olean-12-en-3ß-ol) were identified in petioles and blades of *Kalanchoe blossfeldiana*, in untreated and in methyl jasmonate treated plants (Tab. 3). JA-Me decreased the content of campesterol in petioles and increased the content of ß-sitosterol in blades. It is well known that ß-amyrin is produced in some intact plants, for example in *Pisum sativum* (S u g a and S h i s h i b o r i, 1975) and in callus cultures of *Tylophora indica* and *Rosa* 'Paul's Scarlet' (S t o h s and R o s e n b e r g, 1975).

In the petioles and blades of control plants, and in the petioles of plants treated with JA-Me olean-12-one was identified (Tab. 3). The chemical structure of the compound is similar to ß-amyrin; only one group OH^- is absent in olean-12-one in comparison to ß-amyrin.

It is interesting that only in blades of *Kalanchoe* treated with methyl jasmonate was identified 2H-cyclopropa[a]naphthalen-2-one, 1, 1a, 4, 5, 6, 7, 7a, 7b-octahydro-1, 1, 7, 7a-tetramethyl (aristolone) (Fig. 1, Tab. 3). The sesquiterpene aristolone was identified in *Aristolochia debilis* (N i s h i d a and K u m a z a w a, 1973), in some species of tropical trees and *Porella caespitans* subsp. *setigera* and *P. faurieri* (*Hepaticae*) (A s a k a w a et al., 1980).

Table 3

The effect of methyl jasmonate on the content of sterols and aristolone (µg g^{-1} fresh weight) in petioles and blades of *Kalanchoe blossfeldiana* 1 day after treatment made on (February 7) during full flowering

Compounds	Petioles		Blades	
	Control	JA-Me	Control	JA-Me
ß-Sitosterol	73.3a	61.4a	134.6b	183.0a
Campesterol	16.6a	11.9b	18.7a	26.8a
ß-Amyrin	+	+	+	+
Olean-12-one	+	+	+	–
Aristolone	–	–	–	+

Explanation: + present, – absent

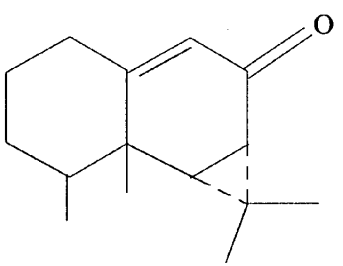

Fig. 1. Chemical structure of aristolone (A s a k a w a et al., 1980)

It is well known that methyl jasmonate greatly induced different secondary plant products specific for various cell cultures of higher plants (Gundlach et al., 1992; Mizukami et al., 1993). JA-Me induced secondary metabolites by a factor of 9-30 over the content values in these systems. The induction by jasmonate does not appear to be specific to any type of secondary metabolites, but rather general to a wide spectrum of low molecular weight substances ranging from flavonoids, guaianolides, and anthraquinones to various classes of alkaloids (Gundlach et al., 1992; Mizukami et al., 1993).

It seems that leaf abscission induced by methyl jasmonate in *Kalanchoe blossfeldiana* is not connected with changes in content of free and bound fatty acids and sterols.

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Zawartość kwasów tłuszczowych i steroli w czasie indukowanego odpadania liści *Kalanchoe blossfeldiana* przez ester metylowy kwasu jasmonowego

Streszczenie

Poprzednio wykazano, że ester metylowy kwasu jasmonowego (JA-Me) indukuje odpadanie liści u *Kalanchoe blossfeldiana*. W obecnych badaniach wykazano, że JA-Me nie wpływa lub w niewielkim stopniu na zawartość wolnych i związanych kwasów tłuszczowych w ogonkach i blaszkach liściowych. ß-Sitosterol, kampesterol i ß-amyrynę zidentyfikowano w ogonkach i blaszkach liściowych *K. blossfeldiana*; JA-Me obniżał zawartość kampesterolu w ogonkach i podwyższał zawartość ß-sitosterolu w blaszkach. W blaszkach liściowych roślin traktowanych JA-Me stwierdzono zanik olean-12-onu a pojawienie się dodatkowo 2H-cyklopropa[a]naftalen-2-on, 1, 1a, 4, 5, 6, 7, 7a, 7b-oktahydro-1, 1, 7, 7a-tetrametylu. Otrzymane wyniki badań są dyskutowane w kontekście indukowanego odpadania liści przez JA-Me u *Kalanchoe blossfeldiana*.