

## Interaction of Methyl Jasmonate and Ethephon in Gum Formation in Tulip Bulbs

Saniewski, Marian  
Research Institute of Pomology and Floriculture

Ueda, Junichi  
College of Integrated Arts and Sciences, Osaka Prefecture University

Miyamoto, Kensuke  
College of Integrated Arts and Sciences, Osaka Prefecture University

Okubo, Hiroshi

他

<https://doi.org/10.5109/4580>

---

出版情報：九州大学大学院農学研究院紀要. 49 (2), pp.207-215, 2004-10-01. Faculty of  
Agriculture, Kyushu University

バージョン：

権利関係：

## Interaction of Methyl Jasmonate and Ethephon in Gum Formation in Tulip Bulbs

Marian SANIEWSKI<sup>1,3\*</sup>, Junichi UEDA<sup>2</sup>, Kensuke MIYAMOTO<sup>2</sup>,  
Hiroshi OKUBO and Jerzy PUCHALSKI<sup>3</sup>

Laboratory of Horticultural Science, Division of Agricultural Botany, Department of  
Plant Resources, Faculty of Agriculture, Kyushu University, 6–10–1 Hakozaki,  
Fukuoka 812–8581, Japan

(Received June 15, 2004 and accepted July 13, 2004)

Gums are induced by *Fusarium oxysporum* f. sp. *tulipae* in tulip bulbs, in which a relatively high level of ethylene is produced. Exogenous ethylene or ethylene-releasing compound (ethephon) also induced gummosis in healthy tulip bulbs but not in other organs, such as stem and leaves. Methyl jasmonate (JA–Me) applied alone as a lanolin paste induced gum formation in tulip bulbs, stem and basal part of leaves. Ethephon applied simultaneously with JA–Me greatly enhanced gum formation in tulip bulbs, independently of the time of the treatment, for example gum production in tulip bulbs treated on September 10, exceeded 153 mg of dry weight with JA–Me 1.0%, 17 mg with ethephon 2.0% and 1206 mg with JA–Me 1.0% + ethephon 2.0%. After about 4 months any gummosis was observed in each treatment. These results suggest that the induction and/or the production of gums are regulated by a signal network of methyl jasmonate and ethylene, especially by their cross talk. Interactions between endogenous jasmonate and ethylene and gene(s) expression responsible for gum biosynthesis are also discussed.

### INTRODUCTION

The phenomenon of gummosis, the process of accumulation and exudation of gum, are widely distributed in the plant kingdom. Gums are a complex of different substances but most important constituents are polysaccharides of highly individual structure (Boothby, 1983). Gums in plants are induced by environmental stress factors such as pathogen infection, insect attack, mechanical and chemical injuries, water stress and others. All of environmental factors are considered to act via ethylene produced in plant tissues (Boothby, 1983).

In tulip bulbs gums were induced by *Fusarium oxysporum* f. sp. *tulipae*, in which a relatively high level of ethylene was produced (Kamerbeek and De Munk, 1976). Exogenous ethylene or ethylene-releasing compound (ethephon) also induced gummosis in healthy tulip bulbs, but not in other tulip organs as stem and leaves (De Hertogh *et al.*, 1980). We have already reported that a new group of plant hormones, jasmonates, has a promoting effect on the induction and/or production of gums in different organs of tulips

<sup>1</sup> Research Institute of Pomology and Floriculture, Pomologiczna 18, 96–100 Skierniewice, Poland

<sup>2</sup> College of Integrated Arts and Sciences, Osaka Prefecture University, 1–1 Gakuen-cho, Sakai, Osaka 599–8531, Japan

<sup>3</sup> Botanical Garden – Center for Biological Diversity Conservation of the Polish Academy of Sciences, Prawdziwka 2, 02–973 Warszawa, Poland

\* Corresponding author (E-mail: msaniew@insad.pl)

(Saniewski and Puchalski, 1988) and stone-fruit trees and their fruits of the Rosaceae (Saniewski *et al.*, 1998a, b; Saniewski *et al.*, 2000b). Methyl jasmonate (JA-Me) exogenously applied as a lanolin paste induced gummosis in bulbs, stem and basal part of leaves of tulips (Saniewski and Puchalski, 1988). It should be mentioned that under natural conditions for normal growth of tulips, gums were not formed in leaves and stems. JA-Me greatly stimulated ethylene production and ACC (1-aminocyclopropane-1-carboxylic acid) oxidase activity in intact tulips (Saniewski, 1989; Saniewski and Wegrzynowicz-Lesiak, 1994, 1995). Although the application of ACC caused an evolution of ethylene much higher than that of JA-Me, ACC did not induce gum formation in stem of tulips. It has been shown, however, that the simultaneous application of ACC with JA-Me greatly accelerates gum formation in stems and leaves of tulip in comparison with JA-Me treatment alone (Saniewski *et al.*, 1998c; Saniewski *et al.*, 2000a). A rapid increase in endogenous levels of jasmonates, mainly JA, has been found in plants or their organs under stress conditions, for example after mechanical wounding, under osmotic stress conditions and after pathogens infection or insect attack (Saniewski, 1997). These facts strongly suggest that jasmonates are important key compounds on the induction and/or the production of gums in plants together with or without endogenous ethylene. Jasmonates has been known as compounds controlling ethylene production in plant tissues as well (Saniewski, 1997). The purpose of this study was to examine the interaction of methyl jasmonate (JA-Me) and ethephon in gum formation in tulip bulbs in different light conditions and at different stages of flower bud development.

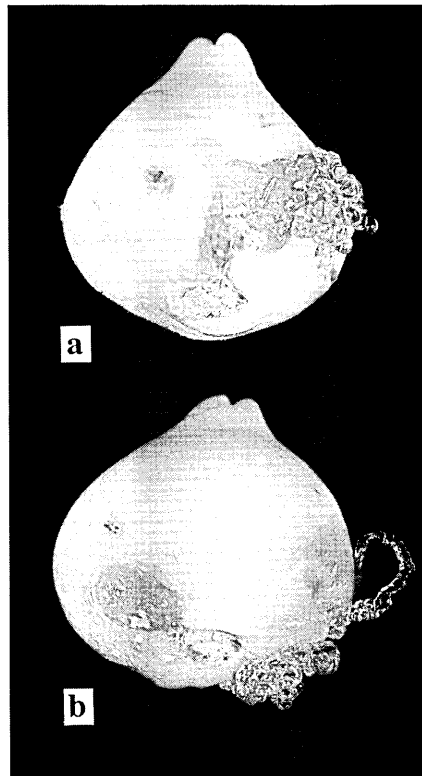
#### MATERIALS AND METHODS

The experiments were performed with the bulbs of tulip (*Tulipa gesneriana* L.) cv. Apeldoorn from commercial stocks. After lifting, the bulbs with circumference of 10–12 cm were stored at 18–22 °C until December 7. Other part of bulbs were cooled at 5 °C starting from October 15. After the removal of dry scales the bulbs were treated around the basal side of scale (about 1.5 cm in width) with JA-Me, ethephon and their mixture in lanolin paste as a ring. A lot of 15 bulbs were used in each treatment. Treated bulbs were stored in greenhouse in natural light conditions or in darkness. Concentrations of JA-Me, ethephon and their mixture used in this study are presented in Figs. 4 and 5. Experiments were carried out in 2000 and 2001 years in different period starting from lifting until December 7. Gums produced by each treatment were weighed one month after treatment.

The data were subjected to an analysis of variance and Duncan's multiple range test at 5% of significance was used for means separation.

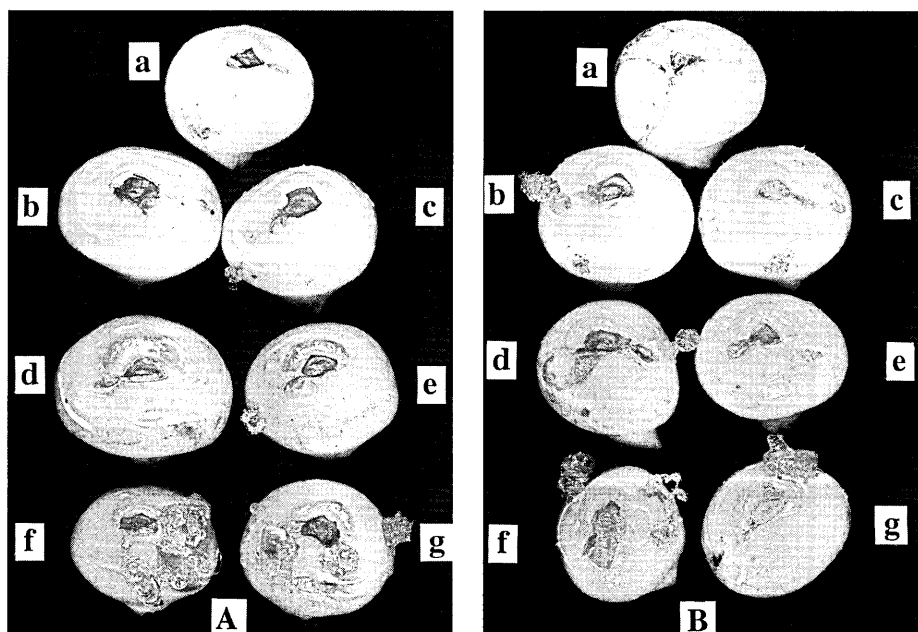
#### RESULTS AND DISCUSSION

Tulip bulbs treated with ethephon and JA-Me, respectively, starting from the beginning to the end of July, induced formation of large amounts of gums (Fig. 1). Extrusion of gums on surface of bulbs started 4–6 days after treatments and then solidified. Starting from August 21 interaction between ethephon and JA-Me at different concentrations was studied. As shown in Figs. 2–5, the abilities of JA-Me and ethephon for



**Fig. 1.** Effect of ethephon (a) and JA-Me (b) at a concentration 1.0% in lanolin paste on gum formation in tulip bulbs cv. Apeldoorn; treatment made on July 21, photographed on August 10.

the induction and the production of gums decreased gradually from August to December. Ethephon applied simultaneously with JA-Me greatly enhanced gum production in bulbs in every date of treatment (Figs. 2–5). This kind of interaction was observed both in natural light conditions and in darkness. However, tulip bulbs treated with ethephon and kept in darkness produced higher amounts of gum than in natural light conditions. Slight gum formation was observed in simultaneous application of JA-Me and ethephon in December although no gums were found in the treatment of JA-Me and ethephon alone at that time. Simultaneous application of ACC and JA-Me has already been reported the acceleration of the onset time and the increment of gum formation in tulip leaves (Saniewski *et al.*, 1999). Maximum gum formation was observed in simultaneous application of JA-Me and ethephon in September, it being over 1200 mg dry of weight. Kamerbeek and De Munk (1976) reported that the occurrence and severity of gummosis were dependent on the stage of tulip bulbs, maximal gummosis took place at 2 to 4 weeks



**Fig. 2.** Effect of ethephon, JA-Me and their interaction, applied in lanolin paste, on gum formation in tulip bulbs cv. Apeldoorn; treatment made on September 6, 2002 and photographed one month later.

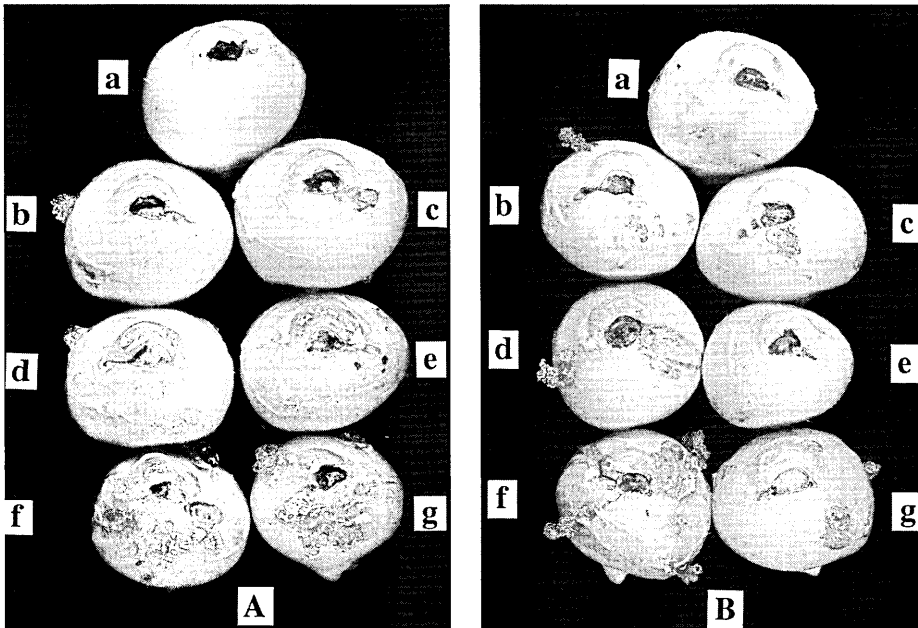
A—after treatment bulbs were stored in greenhouse in natural light conditions (18–24 °C)

B—after treatment bulbs were stored all time in darkness at temperature 20 °C

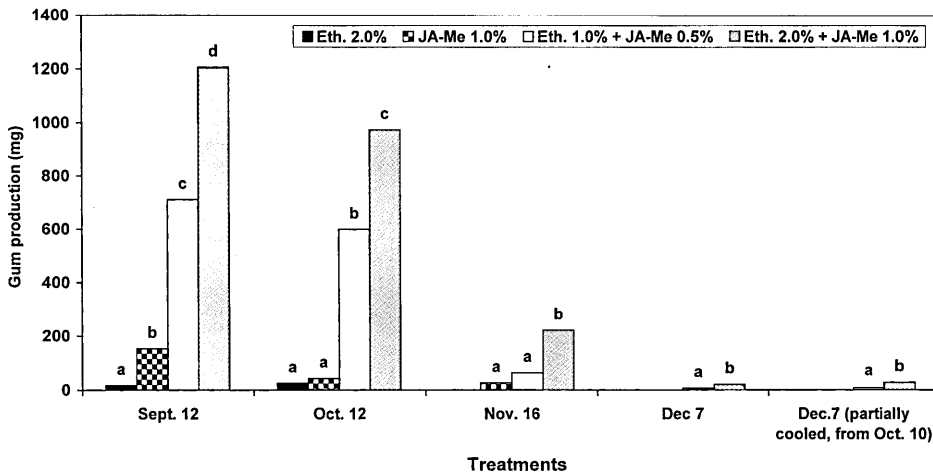
a) control, lanolin paste only, b) ethephon 1.0%, c) ethephon 0.5%, d) JA-Me 1.0%, e) JA-Me 0.5%, f) ethephon 1.0% + JA-Me 1.0%, g) ethephon 0.5% + JA-Me 0.5%.

after lifting and after about 4 months no gums were observed.

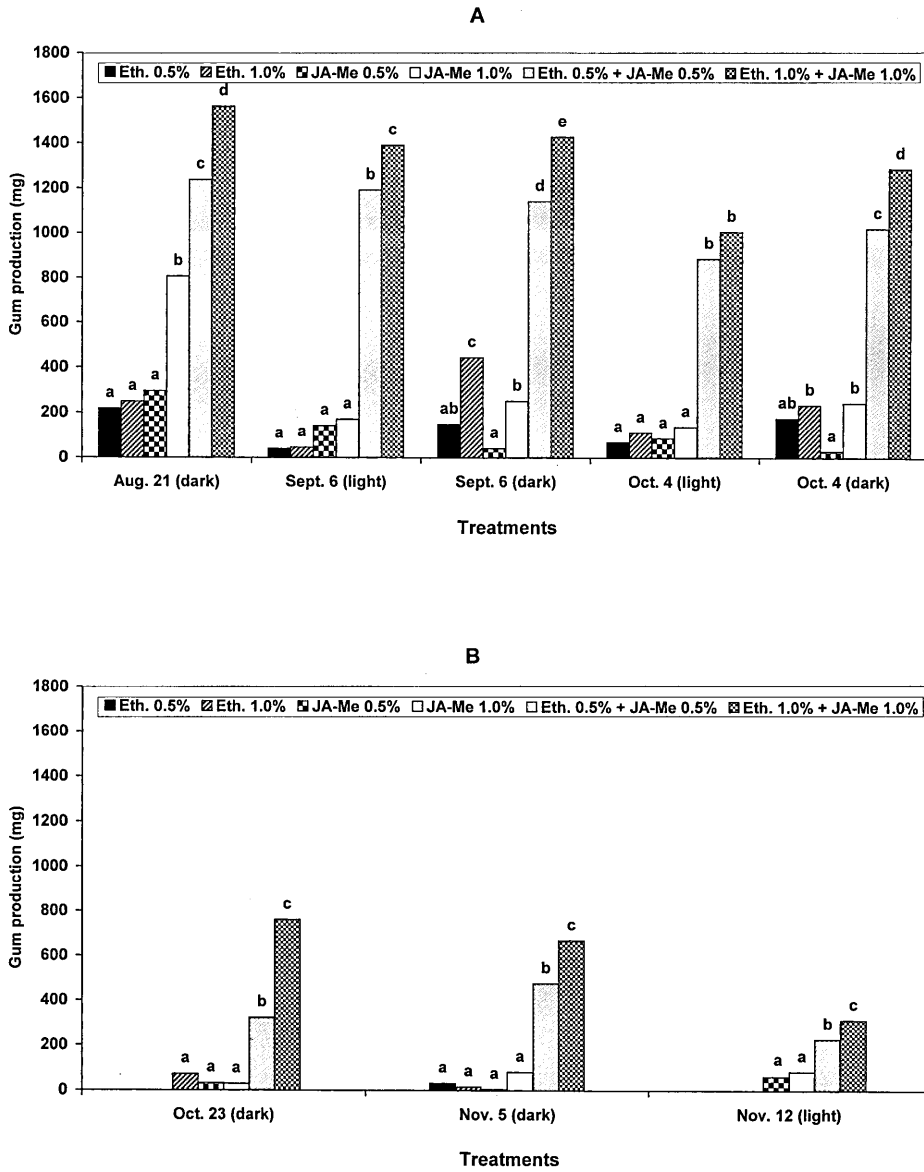
Polysaccharides of gums formed in leaves and stems of tulip induced by simultaneous application of JA-Me and ACC contained ca 16% of putative uronic acid, and remaining neutral sugars consisted of arabinose (ca 40%) and xylose (ca 60%). These results suggest that tulip gums consist of glucuronoarabinoxylan (GlcN:Ara:Xyl=1:2:3) (Saniewski *et al.*, 2000a). The chemical composition of gums induced by ethephon and JA-Me alone in tulip bulbs may be quite similar although it has not been analyzed yet. Chemical composition of polysaccharides (after hydrolysis) in gums of apricot shoots induced by JA-Me as compared with those by ethephon and their mixture has extensively been studied, resulted in the successful identification and quantification of monosaccharides: xylose, arabinose and galactose (1:10:14 at molar ratio) (Saniewski *et al.*, 2001). Uronic acids contents, however, have not been determined yet. These results suggest that the biosynthetic pathway(s) of polysaccharides affected by ethylene and JA-Me is slightly different among plant species. What kinds of carbohydrates (precursors) participate in gum formation induced by JA-Me in leaves, stems and bulbs of tulip and biosynthetic pathway(s) of polysaccharides consisting gums induced by JA-Me are still unknown. Histological



**Fig. 3.** Effect of ethephon, JA-Me and their interaction, applied in lanolin paste, on gum formation in tulip bulbs cv. Apeldoorn; treatment made on October 4, 2001 and photographed one month later. A, B and a-g; see in Fig. 2.



**Fig. 4.** Interaction of ethephon (Eth.) and JA-Me in gum production in tulip bulbs; experiment made in greenhouse in natural light conditions in 2000.



**Fig. 5.** Interaction of ethephon (Eth.) and JA-Me in gum production in tulip bulbs; experiments made in 2001.

studies of tulip stem in different stages in gum formation induced by JA-Me suggest that the degradation products of cell walls and protoplasts contribute to the gum exudates (Saniewski and Dyki, 1997).

One of important questions is why the response of tulip bulbs cv. Apeldoorn for gum formation after infection with *Fusarium oxysporum* f. sp. *tulipae*, ethylene and JA-Me treatment disappears gradually 2 to 4 weeks after lifting. One of the possible explanations is the connection of metabolic changes during flower bud formation including some senescence processes of scales. Another open question is why only some tulip cultivars show a strong gummosis response due to ethylene treatment (for instance cv. Apeldoorn, cv. Enterprise), whereas bulbs of other cultivars (for instance cv. Red Champion, cv. White Sail) show hardly any response to ethylene – lack of gummosis (Kamerbeek *et al.*, 1971). It might depend on the difference of sensitivities of tulip tissues for ethylene and/or JA-Me.

Mechanism of the synergistic interaction between ethephon (ethylene) and JA-Me in gum induction and/or production in tulip bulbs has not been clear yet. The processes of gum induction and/or production may be regulated by a signal network of ethylene and JA-Me, especially by their “cross-talk”. It is possible that in the case of exogenously applied ethylene or JA-Me, interaction between endogenous jasmonate and ethylene, and gene(s) expression responsible for gum biosynthesis takes place (Saniewski *et al.*, 1999). Jasmonates consist of an integral part of the signal transduction chain between stress signal(s) and stress response(s). Cooperative cross-talk among jasmonates and various hormone signals, especially ethylene, occurs in regulation of growth and development and in defense responses against a wide variety of abiotic and biotic agents.

Jasmonates have been well known to induce strongly gene expression of wound- or defense-related tissues when plants are under stresses from mechanical injury and pathogen infection or insect invasion, resulting in the accumulation of proteinase inhibitors (PI) and other pathogenesis-related proteins (O'Donnell *et al.*, 1996; Koiwa *et al.*, 1977; Seo *et al.*, 1997). The interaction of jasmonates with ethylene has been reported, in which these compounds acted together to regulate PI gene expression during wound response. Gene expression of these proteins was inhibited by inhibitors of JA and ethylene biosynthesis (O'Donnell *et al.*, 1996). Combinations of jasmonates and ethylene caused synergistic induction of pathogenesis-related gene expression osmotin mRNA in tobacco seedlings (Xu *et al.*, 1994). It remains to be demonstrated whether ethylene acts upstream or downstream in concert with jasmonates. There was documented synergy between jasmonates and ethylene for the induction of the plant defense gene of *PDF1.2* in *Arabidopsis thaliana* infected by *Alternaria brassicicola* (Penninckx *et al.*, 1998). Conceptually, three different models for the interaction between jasmonates and ethylene have been suggested by Penninckx *et al.* (1998). Model 1 implies that pathogen infection initially stimulates production of ethylene, which subsequently stimulates production of jasmonates, which in turn activates *PDF1.2*. Model 2 implies that pathogen infection initially leads to enhance production of jasmonates, which subsequently triggers elevated production of ethylene, which in turn controls *PDF1.2* expression. Model 3 predicts that pathogen infection results in simultaneous production of ethylene and jasmonates, which are both required for induction of *PDF1.2*. Besides them, there are numerous reports related to the relationships between jasmonates- and/or ethylene-induced gene expres-



sions for their signaling and plant defenses. Further investigations will be required for the analysis of mechanisms of this kind of "cross-talk" on induction and/or production of gums in plants.

The physiological role of gums in plants has not been clear yet. It has been believed that gums have a function in limiting the spread of fungal and bacterial pathogens by isolating the infected tissues (Boothby, 1983). It appears that jasmonates represent an integral part of the signal transduction chain between stress signal(s) and stress response(s), and interact with ethylene in many physiological processes, including gum induction and/or production.

### REFERENCES

- Boothby, D. 1983 Gummosis of stone-fruit trees and their fruits. *J. Sci. Food Agric.*, **34**: 1-7
- De Hertogh, A. A., D. R. Dilley and N. Blakely 1980 Response variation of tulip cultivars to exogenous ethylene. *Acta Hort.*, **109**: 205-210
- Kamerbeek, G. A. and W. J. De Munk 1976 A review of ethylene effects in bulbous plants. *Scientia Hort.*, **4**: 101-115
- Kamerbeek, G. A., A. L. Verlind and J. A. Schipper 1971 Gummosis of tulip bulbs caused by ethylene. *Acta Hort.*, **23** (I): 167-172
- Koiba, H., R. A. Bressan and P. M. Hasegawa 1997 Regulation of protease inhibitors and plant defense. *Trends Plant Sci.*, **2**: 379-384
- O'Donnell, P. J., C. Calvert, R. Atzorn, C. Wasternack, H. M. O. Leyser and D. J. Bowles 1996 Ethylene as a signal mediating the wound response of tomato plants. *Science*, **274**: 1914-1917
- Penninckx, I. A. M. A., B. P. H. J. Thomma, A. Buchala, J.-P. Mettraux and W. F. Broekaert 1998 Concomitant activation of jasmonate and ethylene response pathways is required for induction of a plant defending gene in *Arabidopsis*. *Plant Cell*, **10**: 2103-2113
- Saniewski, M. 1989 Relationship between stimulatory effect of methyl jasmonate on gum formation and ethylene production in tulip stem. *Bull. Pol. Acad. Sci., Ser. Sci. Biol.*, **37**: 55-67
- Saniewski, M. 1997 The role of jasmonates in ethylene biosynthesis. In "Biology and Biotechnology of the Plant Hormone Ethylene", ed. by A. K. Kanellis, C. Chang and D. Grierson, Kluwer Academic Publ., Dordrecht, Boston, London. pp. 39-45
- Saniewski, M. and B. Dyki 1997 Histological changes in tulips stem during gum formation induced by methyl jasmonate. *Acta Hort.*, **430**: 125-131
- Saniewski, M., K. Miyamoto and J. Ueda 1998a Methyl jasmonate induces gums and stimulates anthocyanin accumulation in peach shoots. *J. Plant Growth Regul.*, **17**: 121-124
- Saniewski, M., K. Miyamoto and J. Ueda 1998b Methyl jasmonate induces gums in the shoots of peach and others stone-fruit trees. 16th International Conference on Plant Growth Substances, Makuhari Messe., Chiba, Japan, August 13 to 17, 1998. Abstracts, p. 134
- Saniewski, M., K. Miyamoto and J. Ueda 1998c Gum formation by methyl jasmonate in tulip shoots is stimulated by ethylene. *J. Plant Growth Regul.*, **17**: 179-183
- Saniewski, M. and J. Puchalski 1988 The induction of gum formation in the leaf, stem and bulb by methyl jasmonate in tulips. *Bull. Pol. Acad. Sci., Ser. Sci. Biol.*, **38**: 35-38
- Saniewski, M., J. Ueda, M. Horbowicz, K. Miyamoto and J. Puchalski 2001 Gum induction in apricot (*Prunus armeniaca* L.) shoots induced by methyl jasmonate. *Acta Agrobotanica*, **54**: 27-34
- Saniewski, M., J. Ueda and K. Miyamoto 1999 Interaction of ethylene with jasmonates in the regulation of some physiological processes in plants. In "Biology and Biotechnology of the Plant Hormone Ethylene", ed. by A. K. Kanellis, C. Chang, H. Klee, A. B. Bleecker, J. C. Pech and D. Grierson, Kluwer Academic Publ., Dordrecht, Boston, London, pp. 173-187
- Saniewski, M., J. Ueda, K. Miyamoto and M. Horbowicz 2000a Gum induction by methyl jasmonate in tulip stem. Relevance to its chemical composition. *Acta Hort.*, **515**: 39-48
- Saniewski, M., J. Ueda, K. Miyamoto, M. Horbowicz and J. Puchalski 2000b Methyl jasmonate induces gummosis in plants. *Human and Environmental Sciences*, **9**: 93-100
- Saniewski, M. and E. Wegrzynowicz-Lesiak 1994 Is ethylene responsible for gum formation induced by

- methyl jasmonate in tulip stem? *J. Fruit Ornam. Plant Res.*, **2**: 79–80
- Saniewski, M. and E. Wegrzynowicz-Lesiak 1995 The role of ethylene in methyl jasmonate-induced gum formation in stem of tulips. *Acta Hortic.*, **394**: 305–313
- Seo, S., H. Sano and Y. Ohashi 1997 Jasmonic acid in wound signal transduction pathways. *Physiol. Plant.*, **100**: 740–745
- Xu, Y., P.-F. L. Chang, D. Liu, M. L. Narasimhan, K. G. Raghothama, P. M. Hasegawa and R. A. Bressan 1994 Plant defense genes are synergistically induced by ethylene and methyl jasmonate. *Plant Cell*, **6**: 1077–1085