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## Biochemical interactions between triticale (*Triticosecale*; Poaceae) amines and bird cherry-oat aphid (*Rhopalosiphum padi*; Aphididae)

Cezary Sempruch<sup>a,\*</sup>, Marcin Horbowicz<sup>c</sup>, Ryszard Kosson<sup>b</sup>, Bogumił Leszczyński<sup>a</sup>

<sup>a</sup> Siedlce University of Natural Sciences and Humanities, Department of Biochemistry and Molecular Biology, Prusa 12, 08-110 Siedlce, Poland <sup>b</sup> Research Institute of Horticulture, Konstytucji 3 Maja 1/3, 96-100 Skierniewice, Poland

<sup>c</sup> Siedlce University of Natural Sciences and Humanities, Department of Plant Physiology and Genetics, Prusa 12, 08-110 Siedlce, Poland

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#### ABSTRACT

The polyamines (PAs) and tryptamine are low molecular weight compounds involved in plant responses to abiotic and biotic stresses. The phenomenon is well known in case of pathogenic fungi, bacteria and virus infections. However, their role in plant defence against herbivorous arthropods is not clear. Thus, the quantification of polyamines and tryptamine within tissues of less and more susceptible triticale cultivars (cvs.) to bird cherry-oat aphid (Rhopalosiphum padi L.) was performed. The obtained results showed that shoots of the studied cvs. contained such polyamines as putrescine, cadaverine, spermidine and spermine, and aromatic monoamine tryptamine. More susceptible Tornado cv. was characterised by a higher content of putrescine and spermidine, and a lower level of cadaverine compared to less susceptible Witon cv. The aphid feeding on Tornado seedlings at the beginning caused an increase of tryptamine and spermine content. Prolongation of the infestation reduced the amines level with the exception of spermine after one week of infestation. The less susceptible Witon cv. reacted by increase of putrescine and spermine level, and by decrease of cadaverine and tryptamine content in initial period of infestation. Further extension of R. padi feeding to one week caused reduction of the level of all analysed amines. After two week period the putrescine level was still decreased, while cadaverine, spermidine and tryptamine was enhanced.

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#### 1. Introduction

The plant defence against aphids is composed of local and systemic responses started by reactive oxygen and nitrogen species, systemin, jasmonic acid (JA), ethylene, salicylic acid, abscisic acid and gibberellic acid (Morkunas et al., 2011). Moreover, there are reports suggesting the participation of plant amines in insect–plant interactions (Kusano et al., 2008; Igarashi and Kashiwagi, 2010). In healthy plant tissues, PAs content is tightly regulated in range from hundred micromolars to a few millimolars (Kusano et al., 2007). At higher concentration these biomolecules are dangerous to living cells through induction of programmed cell death (PCD). Induction of PCD is plant responsible for hypersensitive response towards *Diuraphis noxia* (Kurdjumov) (Moloi and van der Westhuizen, 2006). In case of tobacco hypersensitive response against tobacco mosaic virus it was stated that diamine oxidase and polyamine oxidase oxidized putrescine, cadaverine, spermidine and spermine with simultaneous releasing of  $H_2O_2$  (Yoda et al., 2009). During the initial period of infestation, aphids transiently puncture the epidermis, mesophyll and parenchyma cells, causing responses to mechanical damages (Goggin, 2007).

*Abbreviations:* HCAAs, hydroxycinnamic acid amides; JA, jasmonic acid; PAs, polyamines; PCD, programmed cell death; TYDC, tyramine decarboxylase. \* Corresponding author. Tel.: +48 25 6431224; fax: +48 25 6431367.

*E-mail address:* cezar@uph.edu.pl (C. Sempruch).

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Wounding of plant tissues increased the content of free and conjugated PAs and aromatic monoamines as well as induced the activity of key enzymes of their biosynthesis (Groppa and Benavides, 2008). For example, wounded tissues of transgenic tobacco accumulated tyramine-derived hydroxycinnamic acid amides (HCAAs), and the activity of tyramine decarboxylase (TYDC) and tyramine hydroxycinnamoyl transferase was increased (Hagel and Facchini, 2005). Tebayashi et al. (2007) demonstrated that JA treatment of sweet pepper (*Capsicum annuum* L) caused simultaneous accumulation of caffeoyl putrescine and ovipositional deterrence of leafminer *Liriomyza trifolii* (Burgess). Synthetic *p*-coumaroyl putrescine was also characterised by similar deterrent activity. Induction of biosynthesis of *p*-coumaroyl putrescine and dicaffeoyl spermidine in *Nicotiana attenuata* (Torr. ex Wats.) by silencing NaMYB8 transcription factor, reduced the performance of *Manduca sexta* (L.)

and *Spodoptera littoralis* (Boisduval) (Bassard et al., 2010). The plant HCAAs are toxic for insects since they cause paralysis by binding to quisqualte type glutamate receptors on the exoskeletal muscles and block synaptic transmission (Klose et al., 2002). Moreover, aromatic monoamines, such as tyramine and catecholamines, may be toxic for herbivorous insects, similarly to other plant phenolic compounds (Kulma and Szopa, 2007).

In spite of the presented data suggesting that plant amines participate in herbivorous insect-host plant interactions as molecules involved in signalling cascades of responses against herbivores, and/or in wounding effect resulting from insect feeding (Chen et al., 2006), the mechanism of this phenomenon is still not clear. Thus the study was aimed at quantifying the differences in PAs and tryptamine content within tissues of two winter triticale cvs. with different susceptibility to bird cherry-oat aphid (*Rhopalosiphum padi* L.) as well as determining changes in their level during aphid infestation.

#### 2. Material and methods

#### 2.1. Plant material

Two cultivars of winter triticale (*Triticosecale*, Wittm. ex A.Camus): Tornado and Witon, which vary in their susceptibility to bird cherry-oat aphid, were used in the experiments. Seeds of the both cultivars were obtained from Plant Breeding and Acclimatization Institute (IHAR) in Strzelce near Łódź (Poland).

#### 2.2. Aphids

Parthenogenetic individuals of *R. padi* were reared on winter triticale seedlings (Tornado cv.) in climatic chamber at 24 °C at day and 18 °C at night, 70% RH and photoperiod 16L:8D.

#### 2.3. Field experiments

The field experiments were carried out at Agricultural Experimental Station in Zawady near Siedlce (Central – East of Poland;  $52 \circ 03'41''$ N,  $22 \circ 33'22''$  E). The abundance of bird cherry-oat aphid on the triticale was estimated in random block arrangements in three replicates for both studied cultivars. The plot area was 2 m × 9 m, and the distances between plots were 3 m.

The bird cherry-oat aphid density on the studied cvs. was estimated according to the method described earlier by Wratten et al. (1979) and Lykouressis (1984). The observations were carried out from the aphid arrival on the cereals, until its disappearance (G.S.52–88; Tottman and Broad, 1987), in one week intervals. The technique of counting aphids on 50 randomly selected plants, diagonally across the field was applied, and population dynamics of *R. padi* on the studied cvs. was performed.

#### 2.4. Laboratory experiments

Seeds of the studied cultivars were germinated in climatic chamber at 24 °C at day and 18 °C at night, 70% RH and photoperiod 16L:8D. Plants were grown in medium nutrient fine structure compost with sand, in  $8.0 \times 9.5$  cm plastic pots, and regularly watered.

#### 2.4.1. Population tests

The adult apterous females were placed individually on abaxial surface of seven days old seedlings of the triticale cultivars. Seedlings with aphids were isolated with Plexiglass cages with a cheese cloth cover (10 cm  $\times$  30 cm). After 24 h, one nymph was left on each single plant, while the other offspring and the adult were removed. The experiment was run in 25 independent replicates for each studied triticale. Aphids' pre-reproductive period (time from birth until maturity of female) and daily fecundity were estimated. An intrinsic rate of natural increase ( $r_m$ ) and mean time of generation development (T) were calculated using the following equations after Wyatt and White (1977):

$$r_m = 0.738 \frac{\ln M d}{d}$$

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