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What makes plants attractive for a polyphagous bug? Investigations on host plant selection of *Halyomorpha halys* for developing a new plant protection strategy for organic farming

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Abstract

The Brown Marmorated Stink bug (BMSB) Halyomorpha halys (Stål) (Hemiptera: Pentatomidae) is an invasive, polyphagous pest insect native to Eastern Asia that causes serious damage in fruit and vegetable farming. No effective plant protection is available in Germany for organic or integrated farming. It is still unknown how BMSB decides whether a plant is a suitable host for feeding. In order to investigate this, a field monitoring of the bugs on different crops was conducted. When high abundances of BMSB on one specific crop were observed, plant volatiles were collected, analysed, and colour spectra of fruit and leaves were measured. Compounds that were found to be shared among all crops were tested using electroantennography. Identifying olfactory and visual cues are the first steps in finding a potential attractant that could then be used in an innovative capsule based attract-and-kill strategy.

Keywords: *Halyomorpha halys*, host selection, organic farming, push-pull-kill-strategy, semiochemicals

Introduction

The Brown Marmorated Stink bug (BMSB) *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae) is an invasive and polyphagous pest insect. Its area of natural distribution is Eastern Asia, but in recent years it has been introduced to North and South Amerika and multiple European countries (Leskey & Nielsen, 2018). This plant-sucking insect is extremely polyphagous with a range of over 200 different plants from several families. Feeding behaviour on fruit crops results in corky, dry tissue on the feeding sites or can cause deformation and discolouration of the fruit (Acebes-Doria *et al.*, 2016). These damages can lead to high economic losses for farmers in the newly invaded regions, especially for fruit growers (Leskey & Nielsen, 2018; Moore *et al.*, 2019). A high robustness against several insecticides, so that only repeated applications or the use of broad-spectrum insecticides are currently effective, make pest control, especially in integrated and organic farming difficult (Leskey *et al.*, 2018). Thus, a new pest management strategy against this invasive insect is needed for sustainable, eco-friendly, and organic farming.

The aim of the joint project BIOBUG is to develop a new, biotechnical plant protection strategy against BMSB in organic horticulture. It is based on a selective push-pull-kill system, where volatile attractants and repellents will be used in combination with a biological insecticide or microbial antagonist. All components will be encapsulated in biodegradable, sustainable materials.

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To design such a strategy, it is important to understand the host plant recognition of the target insect. Different cues are part of host plant attractiveness and acceptance. For long-distance attraction, visual and olfactory signals, such as volatile organic compounds (VOC), have a high importance for various insect species. It is still unknown how particular cues effect host plant selection of BMSB at certain times of plant and fruit ripening. This study gives a first insight in potentially behaviourally important olfactory and visual cues.

Material and Methods

For field monitoring pheromone-baited traps were placed in commercial orchards with apple, pear, plum or quince trees near Heidelberg and in cherry trees at the experimental field station of the Julius Kühn-Institut Dossenheim. When high abundances of BMSB were observed, fruit and leaves were carefully wrapped in cellophane oven bags (Toppits, Melitta, Minden, Germany) and volatiles were collected from those plant parts for 30 min at a flow rate of 1 I/min (Gross *et al.* 2019). Subsequent chemical analyses were conducted using thermodesorption and gas chromatography coupled with mass spectrometry (GC-MS).

Reflectance of fruit and leaves were measured with a Flame Miniature Spectrometer (Ocean Optics Inc., Duiven, The Netherlands).

Electroantennographic recordings (EAG) were performed with nine volatiles detected in the headspace samples volatiles hexanal, trans-2-hexenal, cis-3-hexenyl-acetate, cis-3-hexenyl-benzoate, decanal, methyl salicylate, geranyl acetone, 6-methyl-5-hepten-2-one.

Results and Discussion

The one-season monitoring showed different abundances of BMSB among the fruit trees over the growing season. Initially, *H. halys* was most abundant on cherry, followed later in season by plum, quince, pear, and apple. On the latter we observed the overall highest abundance. *H. halys* is known to move among different host plant species depending on which fruiting structures are available due to the phenology of the plants (Martinson *et al.*, 2015). This indicated that different developmental stages of the plants were more or less attractive for BMSB, so movement may be triggered by both, fruit phenology and the nutritional need of the bug (Acebes-Doria *et al.*, 2017). To decide which host plant is most suitable, the insects might obtain information based on the VOC profile or the colour of fruits or leaves.

By comparing the VOC profiles among crops, significant differences were determined (PERMANOVA p < 0.001; PERMDISP p = 0.82; *N* (permutations) = 10000). Nevertheless, some compounds were detected in multiple samples of the different species at various stages. These single compounds were then tested with EAG and classified as perceptible by male and female BMSB. Some of these VOCs occur in several plant families, like the identified green leave volatile cis-3-hexenyl acetate. Commonly expressed compounds may play a role in general host plant recognition by such a polyphagous insect. Also, for cis-3-hexenyl acetate or the aldehyde hexanal it is already known that both compounds contribute to host attractiveness (Morrison *et al.*, 2018) and foraging behaviour (Noge, 2009).

Another similarity between the studied crops was the colour spectrum of leaves and fruits. All spectra contained a green component at about 550 nm, plum fruits had an additionally peak in the UV range (250 - 400 nm). *H. halys* has a dichromatic vision based on two distinct green- and UV-sensitive photoreceptors (Egri *et al.*, 2023), so green colour and UV may might act as general visual cues.

Further research should focus on behavioural experiments to find out if single VOCs or mixtures and fruit colour have an effect on BMSB behaviour. Moreover, main compounds of the VOC profile of attractive host plants need to be identified and tested. Information on

attractive fragrances and colouring offers a great potential for developing a highly-selective, cost-saving and environmentally friendly push-pull-kill strategy against this pest insect.

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