Grower Summary

CP 193

Identification of the sex pheromone of *Nesidiocoris tenuis,* a damaging pest of commercial tomato

Annual Report

Project title:	**Identification of the sex pheromone of *Nesidiocoris tenuis,* a damaging pest of commercial tomato**
Project number:	CP 193
Project leader:	Michelle Fountain, NIAB EMR
Report:	Final report, February 2020
Previous report:	None
Key staff:	**NIAB EMR** Michelle Fountain, Celine Silva, James Woodward **NRI** David Hall, Dan Bray, Steven Harte, Dudley Farman **Thanet Earth** Robert James
Location of project:	NIAB EMR Natural Resources Institute, University of Greenwich
Industry Representative:	Robert James, Thanet Earth
Date project commenced:	1 November 2019

DISCLAIMER

While the Agriculture and Horticulture Development Board seeks to ensure that the information contained within this document is accurate at the time of printing, no warranty is given in respect thereof and, to the maximum extent permitted by law the Agriculture and Horticulture Development Board accepts no liability for loss, damage or injury howsoever caused (including that caused by negligence) or suffered directly or indirectly in relation to information and opinions contained in or omitted from this document.

The results and conclusions in this report are based on an investigation conducted over a 4-four-month period. The conditions under which the experiments were carried out and the results have been reported in detail and with accuracy. However, because of the biological nature of the work it must be borne in mind that different circumstances and conditions could produce different results. Therefore, care must be taken with interpretation of the results, especially if they are used as the basis for commercial product recommendations.

AUTHENTICATION

We declare that this work was done under our supervision according to the procedures described herein and that the report represents a true and accurate record of the results obtained.

Michelle Fountain

Deputy Head of the Pest & Pathogen Ecology Department

NIAB EMR

Signature .....MT Fountain ..... Date 24 Feb 2020

[Name]

[Position]

[Organisation]

Signature ..... Date .....

Report authorised by:

[Name]

[Position]

[Organisation]

Signature ..... Date .....

[Name]

[Position]

[Organisation]

Signature ..... Date .....

GROWER SUMMARY

Headline

Two components of the female sex pheromone of the mirid pest of tomatoes, *Nesidiocoris tenuis,* have been identified and traps baited with the synthetic sex pheromone have been shown to be highly attractive to males in commercial glasshouses.

Background

Nesidiocoris tenuis (Reuter) (Heteroptera: Miridae) is a tropical mirid bug. Although *N. tenuis* predates important insect pests, especially whitefly and larvae of *Tuta absoluta*, it also causes damage by feeding on tomato plants (Sanchez 2008), particularly when prey populations decline. Feeding causes necrotic rings on stems and leaf petioles, flower abortion and punctures on fruits (Figure 1, Calvo et al. 2009). In southern European countries, it is used as a biocontrol agent in protected crops, including tomatoes, e.g. Nesidiocoris-System from Biobest and Nesibug from Koppert. *Nesidiocoris tenuis* is considered an invasive pest in the UK and its use as a biocontrol agent is not approved, but the closely related mirid, *Macrolophus pygmaeus* (Rambur) (formerly *M. caliginosus* Wagner), is widely used.

Figure 1. *Nesidiocoris tenuis* feeding causes necrotic rings on stems and leaf petioles.

Nesidiocoris tenuis causes economic losses due to reductions in yield through feeding on trusses (Arnó et al 2009), increased costs for additional crop protection products, disruption of IPM programme and resultant resurgence in pests, labour to monitor and manage the pest resurgence, and cleaning out glasshouses between cropping. *Nesidiocoris tenuis* has become established in glasshouses growing all-year-round tomato crops in some northern European countries (Jacobson, 2019) and is threatening the 180 ha of tomatoes grown in the UK. Control requires application of pesticides incompatible with IPM programmes, leading to resurgence of whitefly populations and associated viruses, and disruption of other biocontrol systems such as the use of predatory mites against spider mites.

This project aimed to identify, synthesise and field test the sex pheromone of *N. tenuis.* This will pave the way for development of pheromone traps for better monitoring of pest numbers

and more efficient use of control agents. The pheromone could potentially also be used for control of *N. tenuis* by mass trapping or mating disruption and hence reduce or avoid the use of conventional pesticides for control on protected tomato crops in the UK.

Summary

In this project, *Nesidiocoris tenuis* was reared through several generations in the laboratory to provide mated and unmated adults for further work (Figure 2). Volatiles were collected from single or groups of mated or unmated females or males. Whole-body extracts of insect compounds were also made. In analyses of volatiles from females by gas chromatography (GC) coupled to electroantennography (EAG) recording from the antennae of male *N. tenuis*, two components were detected that elicited consistent EAG responses (Figure 3). These were identified by GC coupled to mass spectrometry (MS), and the synthesised compounds had identical GC retention times and mass spectra to the natural compounds. These compounds were thus candidate components of the female sex pheromone. The compounds are coded 'A' and 'B' in this extended Grower's Summary due to the consortium needing to protect intellectual property to ensure continued R&D and ultimately commercialisation of the outcomes from this project. An analogue of one of the pheromone components was synthesised and this elicited an EAG response from the antennae of male *N. tenuis.* This compound is a potential pheromone inhibitor or even repellent that may have applications in control of this pest.

Figure 2. Culturing mated and unmated *Nesidiocoris tenuis* to use in laboratory tests

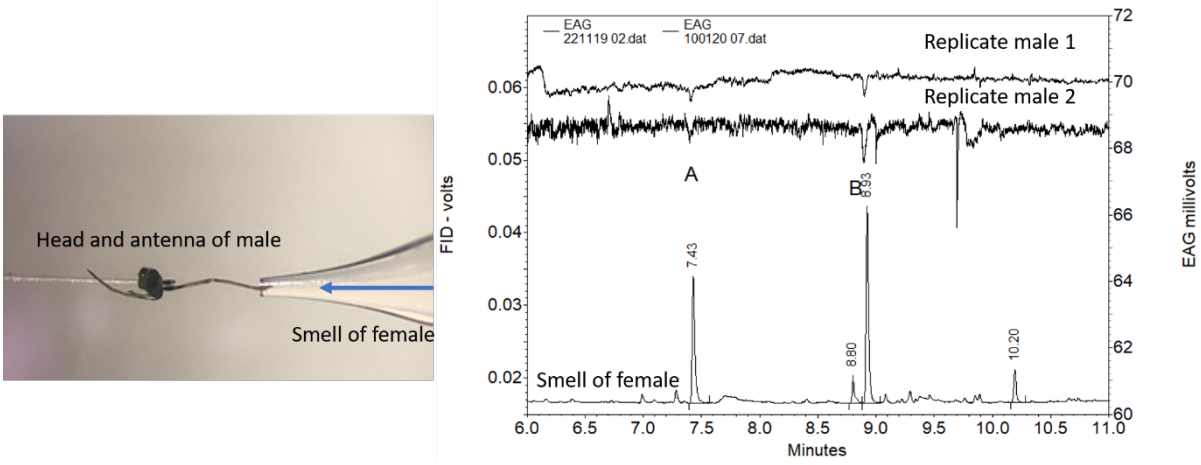

Figure 3. The experimental setup and data output of the GC-EAG. The smell of the female is passed over the head and antenna of the male, which is connected to an electroantennogram (EAG). The components of 'smell' of the female (bottom line of graph) is analysed by Gas Chromatography (GC) and the male antenna response is simultaneously recorded using an EAG (top two lines of graph, representing two replicates). Components of the female smell which triggered a male response are labelled "A" and "B"

Polyethylene vials were shown to be suitable controlled release dispensers for the pheromone components. The release rate could be modified by dilution in sunflower oil and release continued for over two months under laboratory conditions (Figure 4).

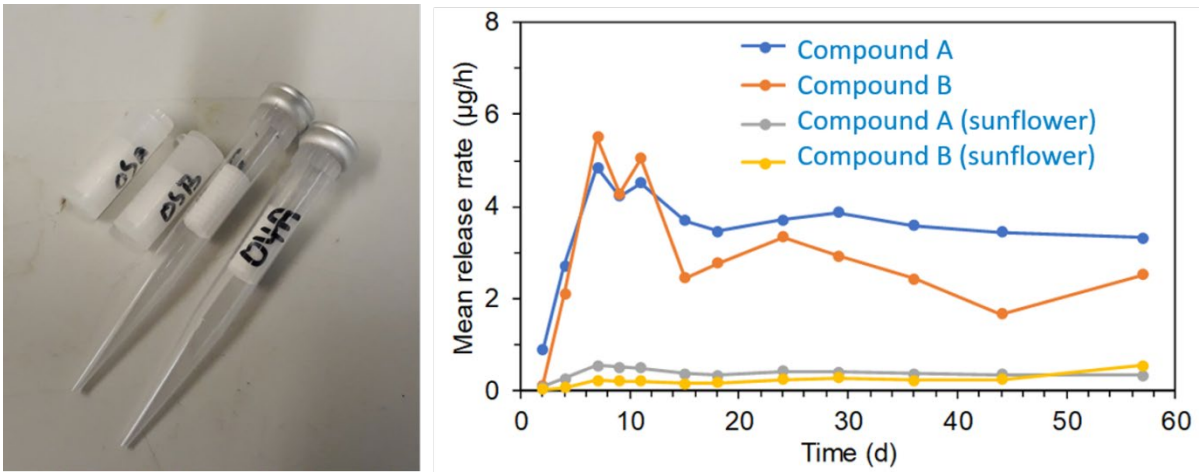

Figure 4. Polyethylene vials and pipette tips (left) used as controlled release dispensers for the pheromone components. The release rates (right) of the 2 compounds from Polyethylene vials, with and without sunflower oil, were measured over time showing that release continued for over two months under laboratory conditions and formulation with sunflower oil modified release rate

Several different laboratory bioassays were trialled, but none showed conclusive attraction of *N. tenuis* males to natural or synthetic sources of pheromone. However, in two experiments carried out in commercial glasshouses, traps baited with a blend of the synthetic pheromone components caught large numbers of male *N. tenuis*. Traps baited with the blend caught significantly more male *N. tenuis* than traps baited with one component, although the latter caught significantly more than unbaited traps (Figure 5). Reducing the release rate of the blend ten-fold reduced catches, but not significantly so. There was some indication that traps at plant height caught more than traps below plant height or significantly above. In discussion with the greenhouse manager the trap catches were thought to provide a good indication of population levels of *N. tenuis* and were greatly reduced following an application of insecticide.

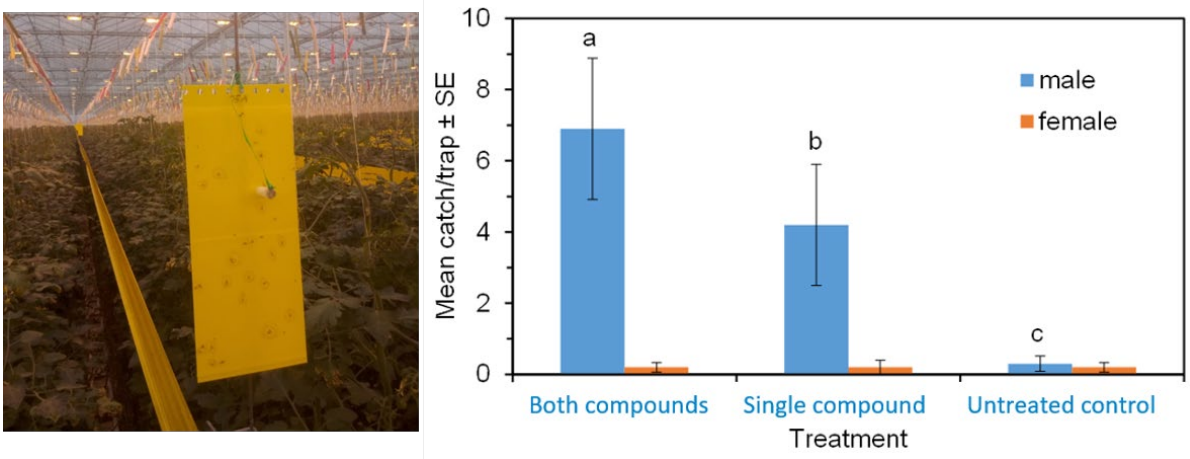

Figure 5. Traps in commercial greenhouse. Traps baited with both compounds caught significantly more male *N. tenuis* than traps baited with a single compound, although the latter caught significantly more than unbaited traps (untreated control). The pheromone was not attractive to females

Although extremely good progress has been made in this short project, further research is required to optimise the blend of pheromone components, the release rate of dispensers, trap design and trap positioning, and then to correlate catches in pheromone traps with population levels and crop damage to develop thresholds for intervention. The use of the pheromone components and the potential inhibitor/repellent in control of *N. tenuis* should also be investigated.

Financial Benefits

If *Nesidiocoris tenuis* became established in UK glasshouses, then additional costs to avoid crop losses estimated by Rob James (Thanet Earth) could be for additional crop protection products (£30k/ha), labour to spot and manage the pests (£10k/ha) and cleaning out glasshouses between cropping (£14k/ha), totalling £54k/ha. If this cost were extrapolated for the 180 ha of tomatoes grown in the UK then this pest could cost the industry approximately £9.7M per crop cycle. Moreover, control in other European countries has required application of pesticides incompatible with IPM programmes or organic programmes, leading to resurgence of whitefly populations and associated viruses, and disruption of other biocontrol systems such as use of predatory mites against spider mites (Jacobson, 2019).

Pheromone traps should provide a means of detecting *N. tenuis* early, more accurately and cost-effectively in order to minimise unnecessary applications of conventional pesticides, thereby reducing costs and disruption of IPM programmes. The pheromone could also be used for control of the pest by mass trapping or mating disruption, which are compatible with IPM programmes. A candidate inhibitor or repellent was also identified during this project and this may also have application in deterring the *N. tenuis* from crops.

Action Points

- Two components of the female sex pheromone of *Nesidiocoris tenuis* have been identified, and traps baited with a blend of the two components have been shown to catch large numbers of *N. tenuis* adult males in commercial greenhouses. Traps and lures are now available for further evaluation by growers and researchers.

Knowledge and Technology Transfer

Fountain MT, Silva C, Woodward J, Hall D, Bray D, Farman D. Identification of the sex pheromone of *Nesidiocorins tenuis*, a damaging pest of commercial tomato. HAPI Meeting 10 Dec 2019.

Fountain MT, Silva C, Woodward J, Hall D, Bray D, Farman D. Identification of the sex pheromone of Nesidiocoris – a damaging pest of tomato. AHDB Protected Edibles Day 2020. Warwickshire, 11 Mar 2020 –cancelled

Fountain MT, Silva C, Woodward J, Hall D, Bray D, Farman D. Identification of the sex pheromone of Nesidiocoris – a damaging pest of tomato. Webinar: Tackling key pests in horticulture, 7th May 2020. https://ahdb.org.uk/events/webinar-tackling-key-pests-in-horticulture

Acknowledgements

The authors are very grateful to Thanet Earth for providing facilities for trap testing and particularly to Janos Dosmodi for setting up and monitoring the traps.

This work was funded by the Biotechnology and Biological Sciences Research Council, a part of UK Research and Innovation, through a joint initiative with the Agriculture and Horticulture Development Board "AHDB/BBSRC Pilot Initiative: Rapid response to high priority pests and diseases in UK Horticulture"

