Grower Summary

CP 082

Discovery and development of new phylloplane biocontrol agents to control insect pests

Annual 2012

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Use of pesticides

Only officially approved pesticides may be used in the UK. Approvals are normally granted only in relation to individual products and for specified uses. It is an offence to use non-approved products or to use approved products in a manner that does not comply with the statutory conditions of use, except where the crop or situation is the subject of an off-label extension of use.

Before using all pesticides check the approval status and conditions of use.

Read the label before use: use pesticides safely.

Further information

If you would like a copy of the full report, please email the HDC office (hdc@hdc.ahdb.org.uk), quoting your HDC number, alternatively contact the HDC at the address below.

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Project Number:	CP 082
Project Title:	Discovery and development of new phylloplane biocontrol agents to control insect pests
Project Leader:	Dr Robert Jackson
Contractor:	University of Reading
Industry Representative:	Dr Neal Ward, Cantello Nurseries Ltd
Report:	Annual Report 2012
Publication Date:	May 2013
Previous report/(s):	N/A
Start Date:	01 October 2011
End Date:	31 December 2014
Project Cost:	£66,150

Headline

Novel aphid-killing biocontrol agents have been discovered that may have use for control of a range of aphids.

Background

Aphid and thrips pests cause major problems in horticulture, through physical damage of crops, deposition of sticky honeydew and the spread of viruses. With a reduction in available pesticides, predatory wasps and *Bacillus thuringiensis* (Bt) bacteria biocontrol agents represent the few options for control, although Bt resistance can manifest rapidly and broadly within pest populations. There is therefore an urgent need for alternative control measures. The aim of this project is to use bioprospecting to identify novel biocontrol bacteria that can kill aphids and thrips and to characterize their efficacy and mode of action. If possible, it would be useful to understand the population dynamics of the bacteria during plant colonization to determine whether certain plants and/or growing conditions would help to proliferate or maintain the biocontrol bacteria. Ultimately, it is hoped that the novel biocontrol bacteria can be commercially developed.

Summary

A semi-targeted approach to isolating and identifying candidate biocontrol bacteria was adopted. It was hypothesized that plants that do not suffer from aphid pests, or have an ability to deter them, might not suffer from these pests due to a component of their microbial microflora. For example, bacteria may occupy the plant surfaces or exist as endophytes i.e. living within the plant tissue. Therefore a range of bacteria were isolated (140 colony types) from eleven different plant species and then inoculated them into a novel aphid-feeding assay to identify bacteria that can kill aphids. From 140 strains tested, nine were found to be effective against six different aphid species: peach potato aphid, *Myzus persicae*, black bean aphid, *Aphis fabae*, cabbage aphid, *Brevicoryne brassicae*, lupin aphid, *Macrosiphum albifrons*, carrot lettuce aphid *Nasonovia ribisnigri* and glasshouse potato aphid *Aulacorthum solani*.

DNA sequence analysis was used to identify the nine bacteria isolated. Although some bacteria (eg *Escherichia fergusonii* and *E. albertii*) were undesirable due them being related to opportunistic human pathogens, most of the bacteria were discovered to be related to harmless environmental bacteria. To focus on the most effective bacteria to use in further experiments, a series of tests were done to discover which strains could be amenable to genetic manipulation and exhibited antibiotic sensitivity – these are key requirements for

identification of toxin and virulence factors. The bacteria were also tested for their ability to kill other insects, an important test of host range as we do not wish to work on bacteria that might kill beneficial insects. Finally, bacteria applied to a surface were tested to establish if they would be ingested by an aphid – this test is important in the context of foliar application.

By assessing dose response and timing of killing, the most potent bacteria that killed the aphids were *Pseudomonas poae*, *Pseudomonas fluorescens* and *Citrobacter werkmanii*. All the bacteria were taken up by aphids from surfaces as well as from liquids, indicating they may be useful for foliar application. These bacteria also demonstrated antibiotic resistance, acceptance of plasmids and the ability to be mutated, which means all would be suitable for genetic manipulation to find the mode of action of aphid killing. To facilitate the studies, DNA of each strain has been sent to the University of Exeter sequencing service to have their genomes sequenced *i.e.* to read the entire genetic blueprint of the bacteria. This will help in our mode of action research.

Finally, contact has been made with a research organisation who have kindly agreed to assist us in screening bacteria against thrips. Results from this should be available within the next six months.

Financial Benefits

Since this project holds more strategic value to gauge the potential for developing novel biocontrol products against aphid and thrips pests, the project remains at an early stage of fundamental science discovery. There is still much work to do in understanding the nature of the aphid-control and thus it is too early at this time to make any recommendations. However, the fact that a range of bacteria that can kill aphids have been discovered suggests it will be possible to develop at least one or more for further characterization and hopefully help in taking them through development and formulation. These would provide growers with a significant financial benefit in reducing losses due to aphids.

Action Points

None to date.