



**UM00055** 

# Ciliate protozoa in baits for the control of grain pest molluscs

# **PROJECT DETAILS**

PROJECT CODE: UM00055

PROJECT TITLE: CILIATE PROTOZOA IN BAITS FOR THE CONTROL OF GRAIN PEST MOLLUSCS

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**END DATE:** 30.06.2016

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#### **Summary**

This proof-of-concept project was aimed at evaluating the potential for using naturally occurring protozoa to control pest molluscs such as the grey field slug (Deroceras reticulatum).

This project aimed to identify effective Australian protozoan parasites of the key pest, grey field slug. Following testing of the capacity for infection of the slugs by ingested protozoa, maternal transmission to eggs and hatchlings was explored. It was shown that adult and hatchling slugs died when challenged with some strains of protozoa. The conditions for the protozoans in the laboratory were determined and the conditions under which they produce desiccation-resistant resting cysts were established.

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#### **Conclusions**

The main conclusions from this research are:

- -Tetrahymena rostrata is present in Australia.
- Australian T. rostrata is amenable to laboratory culture and can be induced to encyst.
- Australian T. rostrata is pathogenic for D. reticulatum and hatchlings are particularly vulnerable.

The challenges for use of *T. rostrata* for biological control of *D. reticulatum* in the field are issues relating to large scale production and determining the best way to deliver viable, infectious ciliates to the slugs and the right time to reduce slug populations following sowing.

The field surveys and research on molecular bar-coding of *Tetrahymena* have conclusively shown that *T. rostrata* is present in a number of geographical locations in Australia. The polymerase chain reaction (PCR) assays can now be used to conduct further field surveys if required and to monitor the spread and survival of ciliates during test release field experiments.

Many specimens of ciliates were collected and some long term, pure cultures were established for further testing. The culture methods can be scaled up with appropriate facilities and expertise.

T. rostrata was induced to encyst simply by starvation and drying, however excystment conditions need to be studied to determine if cysts are a practical form for delivery in baits.

The Australian *T. rostrata* strain was shown to be virulent against slugs and especially hatchlings, thus confirming it could control slug populations in a density-dependent manner provided that there are the right physical conditions of temperature and moisture.

#### Recommendations

This project proved the concept that Australian ciliates can kill slugs. Further research is required on scaling up culture and encystment and investigations on delivery would be required to move forward. The impact on molluscs other than *D. reticulatum* and on beneficial organisms such as earthworms needs to be assessed.

#### **Outcomes**

Slugs cause an average 7.4% yield loss across wheat, barley, oats and canola worth an estimated \$25.9 million p.a. Added to this are current molluscide costs (\$8.7 m) and cultural control costs (\$26.3 m). Snail control costs and losses add a further \$25 m, giving total costs of \$86 m. Current bait technology in field crops is not providing the persistence and efficacy required under the conditions which are ideal for pest activity, even when products are applied optimally before autumn rains and



crop emergence. Resistance may be developing and there are environmental concerns associated with the mis- and over-usage of metaldehyde<sup>#</sup> and methiocarb<sup>#</sup>.

T. rostrata is a globally widespread protozoan which can parasitise and kill the key grain crop pest, the grey field slug and other molluscs, while not posing a health threat to other groups of animals. Tetrahymena species can exist in parasitic and free-living forms and are widespread in Australia. The free-living form can be grown in large numbers on artificial media and induced to form durable cysts, making this protozoan an attractive candidate as a biocontrol agent which could be distributed in baits.

## **Achievements/Benefits**

Field surveys were conducted to determine if ciliates are commonly associated with slugs.

Molecular bar-coding of field-collected ciliates was conducted using PCR assays developed during the project.

Results showed conclusively that *T. rostrata* is present in Australia.

Ciliate culture methods were trialled and long term cultures were established for some isolates.

Bioassays demonstrated that Australian T. rostrata is pathogenic for D. reticulatum and hatchlings are particularly vulnerable.